PASTEUR AND HIS WORK,

FROM AN AGRICULTURAL AND VETERINARY POINT OF VIEW.

BY

GEORGE FLEMING, LL.D., F.R.C.V.S.,
PRINCIPAL VETERINARY SURGEON TO THE ARMY.

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JOURNAL OF THE ROYAL AGRICULTURAL SOCIETY OF ENGLAND.

VOL. XXII.—S.S. PART I.

PASTEUR AND HIS WORK,

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Agriculture, in the widest and most comprehensive sense of the term, depends upon so many collateral sciences, as well as arts, for its continuous prosperity and progressive development, that any marked advancement or important discovery in these must react more or less beneficially upon it, and promote, to a commensurate extent, its welfare. Among the sciences which, for many years, have aided in this direction, perhaps it is not too much to claim for Chemistry a very forward, if not the foremost, place; and among the greatest chemists are those who have devoted at least a portion of their skill and time to the study of what has been termed "Agricultural Chemistry." The chemical composition of soils in relation to the food, growth, and health of plants; the food, feeding, and products of certain animals, as well as the preparation and preservation of many of those products for the use of man; the action of the atmosphere, heat, light, and moisture upon plants and animals; the artificial agents which may be made to second the efforts or supplement the exhausted powers of nature, whether in regard to the soil, the plant, or the animal body—in all these, and in other ways, chemistry has lent its powerful assistance to agricultural requirements, and it may truly be said that, without it, Agriculture would lose one of its best benefactors and most worthy helpmates.

In recent years, the science of Biology has also been bestowing more and more of its favours on Agriculture, and is now pushing Chemistry very hard for the first place in respect to the services it can render the oldest of all the arts, and more especially with regard to the elucidation of the problems which

surround plant and animal life, be that life in a normal or abnormal condition. And here, in mentioning biology, it is impossible to refrain from alluding to the vastly important and wonderful discoveries which have, within half a century, been made through the intelligent employment of, first, the simple, then of the compound microscope. So recently is the date of its most startling revelations, that I myself can remember a rather distinguished Professor of Medicine, not a quarter of a century ago, designating it a scientific toy, and deriding those who, as he expressed it, wasted their time and their eyes in foolishness. This optical instrument is now far more essential to the man of science, and to mankind in general, than the telescope; inasmuch as it reveals to us the presence of the infinitely little —myriads of minute plants and animals, strange organisms and delicate structures—which, until it was employed, were beyond the vision and the knowledge of man, all of which take a part in Nature's work, and many of which have a markedly benignant or malignant *rôle* in the vital operations of the higher plants and animals, they being active agents in the metamorphoses of matter—be it living or dead.

With the introduction of the microscope into biological investigation, a new world in which to make grand conquests has been given to the philosopher and the searcher into life's mysteries; the mysterious phenomena of life and death, growth and decay, building up and breaking down, and even the result of what were supposed to be purely chemical processes, are now within the range of man's scrutiny, and can be ascribed more or less to the operation of the impalpable, and hitherto invisible organisms, the existence of which this optical "toy" has now made us cognisant of. Even "the pestilence which walketh in darkness," destroying man and beast, has been robbed of its mystery by the penetrating light which this ingenious combination of lenses and optical accessories has shed upon it; and man may, by its aid, in time protect himself, and the animals and plants he rears, from disease and destruction, by the knowledge he has thus acquired. Indeed, to some extent this most desirable end has been already achieved; for some diseases, the nature of which was unknown, and in the prevention or cure of which we were simply groping in the dark, are now perfectly understood, and their prevention is based on this understanding; while we are able to make their active cause serve as a protective influence—make, in fact, the bane act as its own antidote—and thus obviate the necessity for resorting to uncertain, oftentimes dangerous, and generally onerous attempts at curing. No greater advance has perhaps ever been

made in the medicine of man and animals, than that which has taken place during this half of the nineteenth century; and to none among those who have contributed to this result is more credit due than to Louis Pasteur, by whom the greatest discoveries in the world of microscopic organisms have been made, the solution of intensely intricate and important problems effected, and the verities of nature—in her darkest and most baffling recesses—demonstrated in a manner which only genius of the highest order could suggest and execute.

It is only too often felt by those who strive to win Nature's secrets, that all the great problems in natural science—such as the nature of heat, of light, of electricity, of gravity—and still more, all questions connected with life, bring us in the end, and frequently after but a few steps, face to face with infinity and mystery. It has been Pasteur's happy lot to select, or rather to be compelled by destiny to follow, a course which has led to such grand achievements, and at every stage of which he has left his indelible and character-mark. His progress has been along the path which has been already trodden by men of great genius, and pursued unfalteringly through weary days and nights, but along which the love of truth burns as a pure and a guiding light—that *lumen siccum* which Bacon insisted should be found in all philosophers, and which, it would seem, neither failure nor disappointment can quench or dim. As a representative of modern science, Pasteur occupies an advanced position. Cicero has somewhere said, "Opinionem commenta delet dies, naturæ judicia confirmat;" and Pasteur in his work appears ever to have borne in mind that speculative opinions have but an ephemeral duration, whilst inferences drawn from nature and truth remain permanently on record.

Originally a chemist, by the force of circumstances and a most fortunate concurrence of events, this most distinguished man became a biologist, and finally a pathologist—startling chemists, physicists, crystallographers, and physicians, no less than agriculturists, with his discoveries, and conferring upon civilisation immediate and inestimable benefits in many directions, while opening up a wide region for the fruitful cultivation of other investigators.

However far-extending and diverse the effects of these discoveries may be, and are, the object of this paper is limited chiefly to a survey of their relations to and influence upon Agriculture, and to a notice of the circumstances and conditions under which they were made, and the benefits likely to accrue from them.

I have stated that Pasteur was originally a chemist; but it may be mentioned that his training in this science was conducted by Dumas at the Sorbonne, and by Balard at the École

Normale, Paris, under whom he became a very competent experimentalist. During his studies in chemistry, molecular physics appear to have proved very attractive to him, and at last to have deeply engaged his attention, the molecular condition of crystals forming the chief object of his investigations. The results arrived at in his inquiry, when only twenty-five years of age, into the symmetrical and unsymmetrical (or dissymmetrical) forms of salts apparently identical in chemical composition, were remarkable, and elicited the admiration of the chief authorities on this subject, and especially of the German chemist, Mitscherlich, who had failed to discover what Pasteur had succeeded in demonstrating, though he had devoted many years to it. The conclusion Pasteur came to was, that the unsymmetrical molecules of matter are produced by, or built up under, the influence of vital agencies, the symmetrical being characteristic of inorganic bodies, the two conditions being typical of the physical barrier that exists between organic and inorganic nature. This conclusion has, however, since been questioned, and Tyndall, twenty years ago, in his 'Fragments of Science,' was inclined to maintain that "it is the compounding, in the organic world, of forces belonging equally to the inorganic, that constitutes the mystery and the miracle of vitality."

It is very probable that had Pasteur continued to pursue his researches in chemistry and molecular physics, he would have attained special eminence, and these sciences would have greatly benefited. At the early age of thirty-two he was appointed Professor of Chemistry at Strasburg, and soon after (in 1854) was transferred to Lille, as Dean of the Faculty of Sciences in that town, where, for a time, he continued to labour, to verify, and to make deductions from theoretic views, until step by step he had discovered the startling connection that existed between his previous researches in chemistry and crystallographic physics, and the new and entirely unexpected results obtained in physiological chemistry—which connection finally led him, as if it were the thread of Ariadne, to his magnificent discoveries in pathology.

This series of successes was referred to by the celebrated Chevreul at the Academy of Sciences some time ago, when he said: "It is by first examining in their chronological origin the investigations of M. Pasteur, and then considering them as a whole, that we are enabled to appreciate the rigour of judgment of that learned man in forming his conclusions, and the perspicacity of a mind which, strong in the truths which it has already discovered, is carried forward to the establishment of new ones."

What might have been considered an accident, led Pasteur

to abandon his hitherto congenial and highly successful line of research in the domain of chemistry and molecular physics, and enter upon a new but not very dissimilar course, in which his great natural gifts and previous training were to confer such advantages. This was the very important study of fermentation, to which his mind was attracted by an almost casual incident while he was at Strasburg.

The observations of a manufacturer of chemicals in Germany had long made it known that the impure tartrate of lime of commerce, if contaminated with organic matters, and allowed to remain dissolved in water during warm weather, fermented, and yielded various products. This excited Pasteur's curiosity, and he prepared some pure right-handed tartrate of ammonia,* to which he added some albuminous matter, and placed the liquid in a warm chamber, where it fermented. During the process of fermentation, the previously limpid mixture gradually became turbid, and the turbidity was found to be due to the presence and multiplication of a microscopic fungus, which, obtaining its sustenance in the liquid, acted as a living ferment.

To the paratartrate of ammonia this mode of fermentation was also applied successfully, the same organism appearing, though there was a wide difference between the results of the two fermentations, so far as the products were concerned; and the important fact was established, that the molecular dissymmetry proper to organic matters intervened in a phenomenon of the physiological order, and did so as a modifier of chemical affinity. The little fungus was able to assimilate the right-handed tartrate more readily than the left, though there was no chemical difference between them—only a difference in molecular constitution. Pasteur was, in this way, the first to introduce into physiological consideration the fact of the influence of the molecular dissymmetry of natural organic products, and in demonstrating that the common mould or mildew could live and multiply on a purely mineral soil, such as the phosphates of potash, of magnesia, and an ammoniacal salt of an organic acid. Sowing the seeds of this mould—*Penicillium glaucum*—in a solution of pure paratartrate of ammonia, it was seen that in germination the left-handed acid appeared in proportion as the right-handed disappeared, the only aliment the plant obtained for its growth being the carbon in the tartaric acid.

These remarkable experiments led Pasteur to infer that ferments were always living organisms, what had previously been looked upon as ferments being merely their food. The yeast-plant had been previously discovered by Leuwenhoeck in Hol-

* A salt which turns the plane of polarised light to the right; there is also a left-handed tartrate of ammonia.

land, Schwann in Germany, and Cagniard-Latour in France; but its real function was not known, and the chemists proclaimed fermentation to be a purely chemical process. The experiments made by Gay-Lussac at the commencement of this century, seemed to them to prove that the oxygen of the air was the *primum movens* in the process; and Liebig, in promulgating this theory, stated that "the ferments are all nitrogenous substances—albumen, fibrine, casein; or the liquids which contain them, as milk, blood, urine—in a state of alteration which they undergo in contact with the air."

The oxygen was the prime agent in breaking up the unstable union between the complex molecules of these substances, and causing a disturbance and transformation of their ultimate particles, resulting in the production of new compounds. Berzelius and Mitscherlich, however, explained the phenomena of fermentation in a different way—the process was one of *catalysis*—the ferment took nothing from, nor did it add anything to, the fermentable matter, and was an albuminoid substance possessing a catalytic force which enabled it to act by its mere presence or contact. Dumas, nevertheless, thought that in the budding of the yeast-cells there should be some clue to the phenomenon of fermentation, as Cagniard-Latour had already surmised in studying the development of the yeast-plant during the metamorphosis of sugar in water. But until Pasteur took up the subject, it was not in any way or anywhere an accepted hypothesis that organisation or life had any influence on the process.

Pasteur's translation to Lille gave the stimulus to the train of thought engendered by the strangeness of the phenomena he had witnessed, in regard to the molecular dissymmetry of the two tartaric acids, and the effect of a microscopic organism upon them, for this had thrown a new light upon the mystery of fermentation. The wonderful part played by such a minute organism could not be an isolated fact, but beyond it there must lie some great general law. It was argued that all that lives must die, and all that is dead must be disintegrated, dissolved, or gasified; the elements which are the substratum of life must enter into new cycles of life. If things were otherwise, the matter of organised beings would encumber the surface of the earth, and the law of the perpetuity of life would be compromised by the gradual exhaustion of its materials. One grand phenomenon presides over this vast work—the phenomenon of fermentation. But this is only a word, and it suggests to the mind simply the internal movements which all organised matter manifests spontaneously after death, without the intervention of the hand of man. What is, then, the cause of the processes of fermentation, of putrefaction, and of slow

combustion? How is the disappearance of the dead body, or of the fallen plant, to be accounted for? What is the explanation of the foaming of the must in the vintage-cask? of dough which, left to itself, rises and becomes sour? of milk, which curdles? of blood, which putrefies? of the heap of straw, which becomes manure? of dead leaves and plants buried in the earth, which are transformed into soil?

The attempts to solve the problem were numerous, but the hypotheses of the chemists were generally accepted, and Pasteur, being a chemist, might be supposed to favour one or other of these. Not so, however. The inductive method of study and research was his guiding star, and the experimental method of proving all things, was the touchstone of the verity of his inductions. "It is the glory of God to conceal a thing," said the wise Hebrew monarch; "but the honour of kings," he added, "is to search out a matter." Kings in the realms of Science are for ever searching out the hidden things which it is the glory of the Creator to conceal, and their discovery but adds to the glory of concealment.

Located in the principal town of the Département du Nord, one of the chief industries of which is the production of alcohol from grain and beet-root, Pasteur resolved to devote his attention to the study of fermentation, not only with a view to solving the problems in connection with it, but also to apply the knowledge gained to a useful purpose. He studied the spontaneous fermentation of milk (lactic fermentation), in which a portion of the sugar is transformed into lactic acid; and, as a consequence, he found himself opposed to the opinion of the few observers who, detecting living organisms in certain fermentations, imagined the presence of these was accidental, and instead of being favourable to the process, was really detrimental to it. He constantly found an extremely minute microscopical living organism, of well-defined form, consisting of little rods, constricted in the middle, and multiplying by dividing across (fission), each portion forming another rod, which soon underwent the same process of division, and so generation after generation of rods was quickly produced. Other inquirers had failed to observe this organism in the lactic fermentation, through imperfect manipulation and faulty preparation of the liquid: they having mixed chalk with the milk in order to keep it neutral, and employed various nitrogenous substances—all of which rendered it impossible to distinguish the ferment. Pasteur happily avoided this cause of error by boil-

Fig. 1.—*Bacterium Lactis (the Lactic Ferment).**

* This figure, and those on pp. 11, 12, 16, and 72, are reproduced from Dr. Klein's book entitled 'Micro-organisms and Disease,' by permission of the publishers, Messrs. Macmillan and Co.

ing a little yeast in some water, filtering the liquid, dissolving in this some sugar, and then dropping carefully into it a minute quantity of fermenting milk, to act as the seed of the ferment in the limpid saccharine solution. Next day the pellucid fluid had become turbid, because of the active fermentation which had been commenced, and as the chalk dissolved, a deposit took place which the microscope showed was composed of germinating rods of the organism jnst alluded to—the lactic ferment. In another experiment, he substituted for the yeast-water a clear decoction of nitrogenous matter, but the ferment invariably appeared in the same manner.

In order to effectually silence the partisans of Liebig's theory, and indeed of all chemists, as to the accidental presence of the organised ferment, the cells of which, they asserted, perished during fermentation and formed lactate of ammonia, Pasteur unremittingly laboured to demonstrate the falsity of the theory, and to prove that not only was there no ammonia formed during alcoholic fermentation, but if it were added it disappeared, entering into the formation of the new cells. Two notable characteristic and crucial experiments he made, which finally settled the dispute. One was with regard to the yeast of beer or alcohol, and the other to the lactic ferment. He introduced into a pure solution of sugar a small quantity of a crystalline salt of ammonia, and some phosphates of potash and magnesia; into this mixture, which, it will be observed, was destitute of albuminoid matter, he sowed an imponderable quantity of yeast—the living cells of the lactic fermentation. The cells thus sown germinated and multiplied, the sugar fermented, and the phosphorus, magnesium, and potassium of the salts united with elements of the sugar of milk, and were in the end converted into lactic acid. A second experiment made with the same skill and care yielded the same results, and demolished the theory of Berzelius and Liebig, which had no longer any foundation. The whole process took place between the sugar aud the ferment germ—a living organism—which owed its growth and multiplication to the nutriment it found in the sugar: fermentation, in short, was simply a phenomenon of nutrition, the ferment contriving to grow upon the sugar and the mineral salts, the remaining portions of these combining to form, in fermenting milk, alcohol and lactic acid, and in the saccharine fermentation alcohol and acetic acid.

The results of his experiments, and the conclusions he arrived at from a consideration of them, Pasteur laid before the Academy of Sciences in 1857.

By the light of his discovery of the lactic ferment, Pasteur soon found a new ferment—the butyric, which has its own special fermentation, resulting in the production of butyric acid. This

organism consists of minute rods, separate, or united in chains of two, three, or more, which reproduce themselves by division, and which have the power of movement—gliding in an undulating manner, and breaking themselves off from each other by this motile faculty. They can be grown, like the other ferments, in fluids containing fermentable substances, in which they will multiply to an almost indefinite extent, their increase marking the

Fig. 2.—*Clostridium Butyricum, or Bacillus Butyricus* (*the Butyric Ferment*).

Some of the spindle-shaped forms include an oval spore.

progress of the butyric fermentation. In studying this organism or vibrio, Pasteur came upon a new and altogether startling peculiarity of these ferments—that they can not only multiply freely without air, but that the presence of air deprives them of life, and stops the fermentation to which they give rise. A stream of pure carbonic acid, so deadly to animals, may be passed through the fluid in which they are growing, without affecting them; but if a current of atmospheric air be substituted for the acid for a brief space, the organisms subside motionless to the bottom, and fermentation is at once arrested.

The question as to the way in which the ferments induced the phenomena of fermentation had to be answered, and in attempting it Pasteur was brought nearer to a solution of the mystery. The micro-organisms, like the higher animals, were nourished upon suitable pabulum—living upon a portion of the fermentable matter; but while the animal, for a given weight of nutritive matter ingested, assimilates a certain quantity, the "microbe" (as Sedillot named these minute plants), in consuming some of the matter, decomposes a quantity far in excess of its own weight. The "must," or sweet wort of beer or wine, when placed in vats or barrels to produce these fluids, will undergo fermentation when yeast has been purposely added, or when the ferment-germs have been accidentally introduced; and the vital actions of the germs — multiplication, and increase in weight and volume—go on entirely independently of the free oxygen of the air, or of that in the "must." In the immense vats of breweries, fermentation disengages quantities of carbonic-acid gas, which is so much heavier than the atmosphere, that it rests in a dense layer on the surface of the fluid, and completely excludes the

air. Yet the ferment-cells multiply with extraordinary rapidity, notwithstanding the entire absence of air or free oxygen; while their activity during this period is exhibited in the enormous difference between their weight, when collected as yeast at the termination of the process, and the weight of the "must" or sugar which has fermented, and been transformed into alcohol, carbonic acid, and some other products. It has been computed that a pound of the ferment will cause the transformation of one hundred and fifty pounds of sugar into alcohol.

Fig. 3.—*Torula or Saccharomyces* (*Yeast Fungus*).

In the lower part of the figure an ascospore and four isolated spores (after Rees) are shown. Magnifying power about 700.

In shallow vessels, Pasteur found that the ferment was even more active than in deep vats, though exposed to the air; but then much less sugar was decomposed—not more than five or six pounds. It was thus demonstrated that the more free oxygen the ferment consumes, the less does it act as a ferment; while the more completely its vital functions are carried on independently of air or free oxygen, the greater is its power of transforming or decomposing sugar. Life without air, and the process of fermentation, are correlative incidents; though it is to be observed that oxygen is essential to the life and growth of the ferment-cell, and when not obtainable from the air, it takes it from the saccharine matter, which contains it; in doing this it produces alcohol, which is merely sugar *minus* some of its oxygen. This ferment-cell or vibrio, and others of its kind, can only live and multiply without air so long as it receives a sufficient quantity of suitable food; when this is consumed, it dies, and further transformation of the matter ceases, unless another kind of organism finds access.

These investigations led Pasteur to recognise two classes of microscopic organisms—one which requires air or free oxygen

in order to exist and multiply, and which he named *ærobies;* and another which could live actively in the absence of free oxygen, possessing itself of this essential element by taking it from its combinations in the food supplied to it—this class he designated *anærobies*, and its discovery caused much astonishment.* Dumas, the celebrated chemist, said one day at the Academy of Sciences, in addressing Pasteur in reference to the last-named class: "In these infinitely small organisms you have discovered a third kingdom — the kingdom to which these organisms belong, which, with all the attributes of animal life, do not require air for their existence, and which find the heat that is necessary for them in the chemical decompositions they set up around them."

The potency of the anærobic class to act as ferments, it may be observed, depends upon their capacity to live without air, by breaking down pre-existing compounds, and forming new and simpler ones; this done, they perish, and then the ærobic germs may, in their turn, live upon them, and convert their remains into other compounds.

Carefully conducted experiments demonstrated that fluid organic matters deprived of all microscopic germs, retained free oxygen for any length of time, and remained unchanged; but if living germs were allowed access to such matters when kept in closed vessels, in a few days there was no oxygen, but carbonic acid. So it was proved that, contrary to the notion previously entertained, oxygen has but little influence in promoting decomposition when germs are absent; though when they are present it acts most powerfully.

Putrefaction is simply fermentation, the sole agent in one as in the other being microscopic organisms, the fermentation of sugar being simply the putrefaction of sugar. It had long been known that fungi, or microscopic animalculæ, were present in putrefying organic compounds; but that they were the real agents in effecting putrefaction was not proved, and by such authorities as Liebig was even denied. Here, again, Pasteur showed that the destruction of animal and vegetable matter was a process of slow combustion, brought about by appropriation of oxygen from the air, through the instrumentality of the ærobies, which, in reality, have the faculty of consuming the oxygen, and are the powerful agents in restoring to the atmosphere and the soil the elements of things which have lived. Mildew, mould, and other cell-formations, two thousand of which would not measure a millimetre, carry on the great work of maintaining the equilibrium between life and death—they themselves dying and

* I venture to express my idea that the bearing of this discovery upon the phenomena attending the production of sweet and sour ensilage respectively may be worthy of careful investigation.—Edit.

being preyed upon by others; so that the ferments are fermented by other ferments.

"In the destruction of that which has lived," says M. Valery Radot, in his 'Life of Pasteur,' "all reduces itself to the simultaneous action of these three great natural phenomena—fermentation, putrefaction, and slow combustion. A living organism dies—animal or plant, or the remains of one or the other. It is exposed to the contact of the air. To the life which has quitted it, succeeds life under other forms. In the superficial parts, which the air can reach, the germs of the infinitely small ærobies hatch and multiply themselves. The carbon, the hydrogen, and the nitrogen of the organic matters are transformed by the oxygen of the air, and under the influence of the life of these ærobies, into carbonic acid, vapour of water, and ammonia-gas. As long as organic matter and air are present, these combustions will continue. While these superficial combustions are going on, fermentation and putrefaction are doing their work in the interior of the mass, by the developed germs of the anærobies, which not only do not require oxygen for their life, but which oxygen actually kills. Little by little, at length, by this work of fermentation and slow combustion, the phenomenon is accomplished. Whether in the free atmosphere, or under the earth, which is always more or less impregnated with air—all animal and vegetable matters end by disappearing. To arrest these phenomena, an extremely low temperature is required. It is thus that, in the ice of the Polar regions, antediluvian elephants have been found perfectly intact. The microscopic organisms could not live in so cold a temperature. These facts still further strengthen all the new ideas as to the important part performed by these infinitely small organisms, which are, in fact, the masters of the world. If we could suppress their work, which is always going on, the surface of the globe, encumbered with organic matters, would soon become uninhabitable."

In the results of his researches into the acetic fermentation, Pasteur has not only again shown the uncertainty of mere observation as compared with the reliability of experimentation, especially when such a master institutes decisive experiments that are in conformity with and explain facts, but he has conferred a great benefit upon industries closely allied with agriculture, and has more or less directly benefited agriculture itself. And in these researches he has once more upset the theories of such chemists as Berzelius, Mitscherlich, Liebig, and others, and placed our knowledge of an important industrial process—the production of acetic acid—on a safe and solid basis, by proving that this depends upon the fixation of atmospheric oxygen by a micro-organism.

The manufacture of vinegar is largely carried on at Orleans, being one of the staple industries of that city, and to this Pasteur devoted his attention. When wine becomes sour—as in bottles to which, through faulty corking, air has obtained access —it is noticed that the oxygen which was originally in it has disappeared, and that nitrogen has replaced it; while the alcohol has also vanished, and in its stead is acetic acid or vinegar. The presence of air is necessary for this change, and before Pasteur took up the subject, the ordinary method of manufacturing vinegar was to expose barrels half-full of vinous fluid to the air, and at a certain temperature; the acetic fermentation was set up and carried on, and every week a small quantity of vinegar was drawn from the barrel and an equal amount of new wine added. This was a slow and a somewhat unsatisfactory process, inasmuch as some months elapsed before fermentation was fully established. The notion was that the alcohol in the fermenting wine was changed into vinegar, by the chemical influence of the oxygen in the air, acting in the presence of dead albuminoid matter. But Pasteur proved that this matter had no influence in the change; for though alcohol be mixed with pure water until it is reduced to the strength of wine, and exposed to the air, it will show no trace of vinegar; yet if some wine be put in a bottle, and this be sealed and then heated to about 140° Fahr., the same result will be noted; while if it be not heated it will become sour. More than this, if the bottle which has been subjected to the high temperature, be afterwards opened and the air admitted, the wine sours. In addition to this demonstration, and to still further show that the dead albuminoid matter of the chemists had nothing to do with the conversion of alcohol into acetic acid, Pasteur removed all traces of albuminous substance from wine, and introduced to it a small quantity of phosphates of ammonia, potash, and magnesia; the vessel was then sealed up and laid by for some time. When again examined, the alcohol had become vinegar.

The real cause of the change was the presence of a minute fungus—the *Mycoderma aceti*—known for generations as the "flower of vinegar," which is always seen on the surface of wine undergoing the change, but which Liebig, who knew it, considered a mere coincidence. The formation of vinegar is always preceded by the development, on the surface of the wine, of this very minute plant, which, when magnified, appears as an extremely fine constricted body; its accumulation constitutes sometimes a scarcely perceptible scum, at other times a thin wrinkled film, unctuous to the touch, because of the various fatty matters it contains. It has the singular property of condensing considerable quantities of oxygen, and of fixing this gas upon the alcohol, thereby transforming it into acetic acid. But like the

larger members of the vegetable kingdom, it must have its appropriate aliment, and wine offers this in abundance, in the form of nitrogenous matters and phosphates of magnesia and potash.

To make vinegar from wine, all that is needed is to mix it with one-fourth of its volume of vinegar, and sow on its surface a few seeds of the fungus, which is done by transferring a little of the mycodermic film from a liquid covered with it. If it be summer, or if the room be heated (for it thrives best in the warmth), in at most forty-eight hours the whole liquid is covered by it, and after some days the alcohol has become acetic acid. Pasteur, wishing to give an idea of the prodigious activity and prolificacy of the little organism, stated during a discussion at the Academy of Sciences, that he would undertake in twenty-four hours to cover with it a surface of vinous liquid as large as the hall in which they were assembled, having the previous day sown in it the almost invisible particles of newly-formed

Fig. 4.—*Saccharomyces Mycoderma, or Mycoderma Vini.*

From an artificial cultivation of dilute flourishing material.

d. Branched mycelium. *f α.* Torula stage. *f β.* Mycelial stage.
(After Grawitz.)

Mycoderma aceti. Millions upon millions of the organism spring into existence in twenty-four hours. Nothing is more simple than to obtain it in the first instance, it being one of those so-called "spontaneous" productions which are almost certain to appear on liquids or infusions which contain its necessary food. It is present in the air of towns and buildings, and in wine, vinegar, and other fluids; and if it is desired to procure some of the mycoderma, it is only necessary to place a mixture of wine and vinegar in a warm place, when in a few days there will appear

little greyish patches on the surface, which go on increasing progressively and rapidly. This is the Mycoderma grown from the seeds, or "germs" which the wine or vinegar accidentally contained, or which the air carried,—just as weeds grow in fields from seeds which are brought there by the wind or animals. That the latter may be also instrumental in extending the process of acetic fermentation is beyond doubt, for in vinegar manufactories, and in places where vegetable matter is souring, there are usually seen numbers of little reddish flies which come from we know not where, but which, by means of their feet or probosces, convey the seeds of this cryptogam.

When a bottle containing wine is subjected to a temperature of 120° to 140° Fahr., the wine remains sweet, because the Mycoderma germs in it, and in the air in the bottle, are killed by the heat; but if the bottle be opened, the wine will sour, because new germs gain access. Wine in well-filled bottles, laid flat, does not become acid, because the Mycoderma cannot grow without a sufficient supply of oxygen; for although air penetrates the pores of the cork, yet it is in such minute quantity that the oxidisable constituents of the wine absorb it, without leaving any for the germs in the bottle. But when the bottles are placed upright, the corks become dry, and the air more readily passes through them; so that the germs on the surface of the wine are surrounded by air, multiply, and break up the alcohol into acetic acid.

The Orleans method of manufacturing vinegar, Pasteur determined to improve, according to the discoveries he had made in acetic fermentation. The disadvantages attending the system hitherto were very serious, and have been briefly alluded to, Into large barrels is first introduced a quantity of good vinegar, with about a fiftieth part of wine. Eight days after, some wine is added, and in a week another quantity, until the barrel is half filled. Then vinegar is drawn off to a certain amount, and fresh wine added to replace it, this process being repeated every eight days, the maximum quantity of vinegar obtained every week from one of these casks, or "mothers," as they are called, being a little more than two gallons; but when the casks work badly, which is frequent, this quantity is diminished. It requires three or four months to prepare a "mother," which has to be very regularly fed with fresh wine, or all will be spoiled, and the business must be commenced anew; the manufacture has to go on at all times, whether vinegar be required or not; and the barrels cannot be stirred from their places during the process.

Instead of the "mothers," Pasteur recommended vats placed in a chamber heated to about 76° Fahr., and filled with vinegar and wine, on the surface of which the Mycoderma was sown.

This simple process, founded on the exact knowledge of the cause of acetic fermentation, has been eminently successful. A large manufacturer of Orleans stated, that at the end of a week or ten days all the acetified wine is converted into vinegar, and that from a hundred litres of the former he drew off ninety-five of the latter. After the great rise of the temperature noticed when the vinegar is being formed—due to the combination of the oxygen with the alcohol, that fluid is allowed to cool, is drawn from the vat, barrelled, refined, and is ready for use. The vat being emptied, is cleaned, again charged, the acetified wine sown with the plant, and the same process gone through.

It has long been noticed that vinegar, when kept for some time, becomes turbid and impoverished in a remarkable manner, and finally becomes putrid. Pasteur pointed out the cause of this, and also the remedy. After the alcohol has become changed into acetic acid, the mycoderm still exists, as it can live upon the acid—beginning with the ethereal and aromatic portion, the most valuable—transforming it into carbonic acid and water, and leaving a small quantity of mineral salts and albuminous matter—the decomposed remains of the plant. This neutral organic fluid is a suitable home for moulds and putrefactive organisms, which consequently rapidly grow, the moulds forming a film over the mass beneath, in which anærobic organisms can consume the dead mycoderms; and thus we have putrefaction in the deeper parts, and combustion at the surface. Minute eel-like organisms also appear in vinegar, and rapidly deteriorate it. It is asserted that there is not a barrel of vinegar manufactured on the now obsolete Orleans system which does not contain them in immense numbers, and, astonishing to mention, they were, previous to Pasteur's investigations, actually considered necessary to the production of vinegar. The mischief wrought by these microscopic creatures is owing to their requiring air to live—like the Mycoderma, they are ærobic; and when the vinegar reaches a certain depth, they form a moving stratum in the upper part of the liquid, where they can obtain air. Here, however, they come into competition with the mycoderms for the essential oxygen which they both must have, and there ensues a struggle for existence. If, for some reason, the film of mycoderms is not formed, or its production is delayed, the ever-moving little eels take possession of the surface of the vinegar and absorb all the oxygen; consequently, the mycoderm cannot develop, or it dies. But if acetification is very active, and the plant has occupied the upper strata, the eels are gradually driven away, and take refuge against the moist sides of the vessel, where they compose a thick grey lining, which is all in movement, and where their enemy cannot so seriously injure

them, since they are surrounded with air. Deeper in the fluid they would perish, and they only linger at the sides of the barrel until they get an opportunity to again contend with their vegetable enemy. Pasteur's intervention removed the evil: the vats are thoroughly and frequently cleaned, so that the organisms have no time to do any harm.

Guided by his studies on vinegar, Pasteur has been able to effect great improvements in the manufacture of beer and wine, by which production is cheapened, and the keeping properties of the liquids much enhanced.

These improvements are founded upon the observed injurious effects of the organisms which give rise to the acetic, lactic, and butyric fermentations; and the measures adopted to prevent them are most simple and effective—the process now being known as "Pasteurisation." With regard to beer, he recommended that it should be bottled when fermentation is nearly completed, and the bottles then subjected to a temperature of between 122° and 131° Fahr., so as to destroy all injurious germs. The wort, while cooling, was also to be guarded against all atmospheric germs, and the leaven employed for making it was likewise to be free from them. Wine has its own peculiar micro-organism—the *Mycoderma vini*—which feeds on new wine, but dies as this becomes old. The vinegar ferment cannot live upon new wine, but as soon as the *Mycoderma vini* perishes and decays, the *M. aceti* attacks it and grows rapidly, so that the wine becomes sour. "Flat" wine, and "greasy" wine (peculiar to the white wines of the Loire basin), as well as the "bitterness" of Burgundy wines, are also due to particular microscopical organisms. The ageing of wine mainly depends on its oxidation, the oxygen which was previously mixed in a mechanical manner with it becoming chemically incorporated in it; for new wine, when destitute of air, does not age, and the difficulty in managing wine is to permit a certain amount of air to be present without any deteriorating germs. M. Radot, in alluding to this subject, says:—"In short, according to Pasteur's observations, the deterioration of wines should not in any case be attributed to a natural working of the constituents of the wine, proceeding from a sort of interior spontaneous movement, which would only be affected by variations of temperature or atmospheric pressure; they are, on the contrary, exclusively dependent on microscopic organisms, the germs of which exist in the wine from the moment of the original fermentation which gave it birth. What vast multitudes of germs of every kind must there not be introduced into every vintage-tub! What modifications do we not meet with in the leaves and in the fruit of each individual spoilt vine! How numerous are the varieties of organic

dust to be found on the stems of the bunches, on the surface of the grapes, on the implements of the grape-gatherers! What varieties of moulds and mildews! A vast proportion of these germs are evidently sterilised by the wine, the composition of which, being at the same time acid, alcoholic, and destitute of air, is so little favourable to life. But is it to be wondered at that some of these exterior germs—so numerous, and possessing in a more or less marked degree the anærobic character—should find, at certain moments in the state of the wine, the proper conditions for their existence and multiplication?"

To protect the wines from these injurious organisms, Pasteur demonstrated that it was only necessary to heat them, when bottled, to a temperature of 140° Fahr. for a few moments, in a water-bath. This insures the future soundness of the wine. After having shown the causes which determine the alterations in wine, by introducing a means of practically neutralising them, Pasteur solved one of the greatest economic questions with regard to this industry. By the application of heat, and without injuring their colour or flavour, the limpidity of all wines was guaranteed, while their indefinite preservation was certain if kept in well-closed bottles, or in barrels, even if transported all over the world.

An amusing incident is related in connection with this discovery. Those most concerned in the preservation of wine were at first incredulous as to the heating process not damaging its taste, colour, or limpidness; and Pasteur addressed himself first to wine-merchants and others who were skilled in the detection of alterations in it, with a view to obtain a decisive opinion—for the public had already shown a preference for his heated wine; and at last he organised a large tasting Commission, appointed by the wholesale wine-merchants of Paris. This body, at its first meeting, could not agree as to the superiority of the heated or unheated wines placed before them, many of them thinking the latter had a better flavour than the former; and Pasteur, fancying that prejudice had much to do in influencing them, intimated that at the next meeting there would be no indication as to which was the heated and the unprepared wine, but their palates should alone distinguish them. On that occasion, he offered them samples taken from the same bottle, and as might be expected, there were preferences for one and for the other, the experts not knowing they were from the same source. The Commission, alluding to this experiment, candidly confessed that the differences between the heated and non-heated wines were imperceptible, if they existed, and that the imagination was not without considerable influence in wine-tasting.

The researches of Pasteur had revealed a world of organisms,

whose minuteness had hitherto either enabled them to escape observation, or to conceal their special function in the economy of nature; and the origin of these wonderful living particles, whose operations are so vast and important in their results, could not but arrest attention. Indeed, the question of spontaneous generation, upon which grave issues in pathology in particular depended, was one which Pasteur was in a manner compelled to take up. It was certainly one that had come down to our own day from hoary antiquity, but it was being debated with unusual warmth while he was successfully unravelling mysterious processes, which he traced to the action of microscopical germs, whose source might be ascribed to a spontaneous or fortuitous combination of elements. Aristotle was of opinion that all damp bodies which become dry, and dry ones which become damp, engender animal life; Virgil thought bees were produced from the putrefied intestines of a young bull; and, much nearer our own time, Van Helmont stated that the smells that rise from marshes produce frogs, leeches, slugs, &c.,—nay, he had even the temerity to assert that mice could be produced by keeping a dirty shirt in the mouth of a vessel containing a little corn, which is transformed into these creatures after a number of days—he had witnessed it! and scorpions could be developed from crushed herbs placed in a hole in a brick! In the last century, Needham maintained the doctrine of spontaneous generation, but Spallanzani opposed it; Redi, an Italian naturalist, showed that maggots are not spontaneously developed in meat, but come from the eggs of flies. The introduction of the microscope was seized upon by the "Spontaneists" to support their notions, as in no way could the appearance of animalculæ in previously barren fluids be accounted for. Mistakes might have been made with regard to the origin of mice and maggots, but it could not be so in the case of microscopic living things. How, except by spontaneous generation, could the presence and rapid multiplication of these in decomposing animal or vegetable substances be explained? Buffon even lent himself to this doctrine, and devised a system in explanation of the hypothesis. In 1858, Pouchet, Director of the Museum of Natural History at Rouen, declared before the Academy of Sciences that he had succeeded in demonstrating, in an absolutely certain manner, the existence of certain microscopic living organisms which had been developed without pre-existing germs.

In a series of ingenious and ably-conducted experiments, Pasteur demolished, one after another, the arguments of Pouchet and the other heterogenists, by convincing demonstrations. "There is not one circumstance known at the present day," he

exclaimed in a discourse at the Sorbonne, "which justifies the assertion that microscopic organisms come into the world without germs, or without parents like themselves. Those who maintain the contrary have been the dupes of illusions and of badly conducted experiments, tainted with errors which they knew not how to perceive or avoid. Spontaneous generation is a chimera."

And Flourens, permanent Secretary of the Academy, hitherto neutral in the discussion, said on the same occasion: "As long as my opinion was not formed I had nothing to say; now it *is* formed, I can speak. The experiments are decisive. If spontaneous generation be a fact, what is necessary for the production of animalculæ? Air and putrescible liquids. Now Pasteur puts together air and putrescible liquids, and nothing is produced. Spontaneous generation, then, has no existence. Those who still doubt have failed to grasp the question." Subsequently, in England, Dr. Bastian became the strenuous advocate of spontaneous generation, but the crucial experiments and absolutely convincing demonstrations of Professor Tyndall, finally abolished the erroneous ideas which had prevailed for so many centuries.

As is well known, the production of silk forms the principal industry of several Departments in the South of France, and the rearing of silkworms occupies the time and attention of great numbers of people—chiefly agriculturists. Previous to 1849, this industry had been particularly flourishing; but in that year, after an exceptionally good silk-harvest, and without any appreciable cause, several of the large establishments were visited by disease among the worms, and this in the course of time assumed the proportions of a plague among the silkworm-nurseries, until at last the silk-husbandry of France was on the verge of ruin. The symptoms of the disease were numerous and variable, and sometimes the worms died early, at other times not before the first, second, or third moulting; oftentimes the eggs were sterile. Instead of becoming white, the worms retained a rusty tint; they did not eat; spots appeared on their bodies like black bruises, which were scattered over the head, rings, and feet. Every batch or brood attacked perished. Fresh eggs were imported from abroad, and at first these hatched well—so much so, that the year 1853, when a large quantity of these foreign worms were reared, was estimated as one of the most productive of the century, 130,000,000 francs being derived as revenue from the cocoons. But the following year the eggs from these worms were found to be no better than the French eggs—they were also infected. To add to the misfortune, the malady extended to Spain and Italy, then to Greece and Turkey, until, in 1864,

all the cultivations from every part of Europe were either diseased or suspected of being so; and throughout the extreme East, Japan only was exempt. The plague had followed the trade in silkworm eggs, just as cattle diseases have followed the trade in cattle.

In 1865, the weight of cocoons had fallen so low, that the French revenue sustained a loss of 100,000,000 francs, and the silk-cultivating Departments were in despair. Agricultural and scientific societies, municipal bodies and governments, were all seriously engaged in attempting to discover the cause and a remedy. And there was no lack of hypotheses, suggestions, and cures; while scores of pamphlets upon the malady were published every year, and experiments were undertaken to elucidate the mysterious scourge, and limit its ravages.

The disease was known as "pébrine," owing to the peppered appearance of the skin of diseased worms.

In 1865, in response to a petition signed by 3600 mayors, municipal councillors, and capitalists of the severely-visited Departments, the French Government appointed a Commission to investigate the malady, and Dumas was selected as its Chairman or reporter, because of his great scientific reputation and his personal interest in one of the afflicted Departments. While preparing his report, it occurred to him that Pasteur was the man best fitted to carry out investigations as to the measures required to combat the plague. But Pasteur at first declined to undertake such a heavy task, inasmuch as his success in the enquiry into organised ferments, in their relation to the manufacture of vinegar and diseases of wines, had opened prospects of a prosperous career—in fact, it was at the moment when, disposing of the vexed question of spontaneous generation, the "infinitely little" had become to him, and to science, the "infinitely great." He saw living ferments everywhere, either as the active agents in decomposition or in producing contagious disorders. To forsake a course which he had so fruitfully pursued and made his own, with all its prospective advantages, and to enter upon another which was novel to him, and the determination of which might be the reverse of satisfactory, appeared to be too much of a sacrifice. Dumas appealed to his friendship and his patriotism. "But consider," said Pasteur, "that I have never handled a silkworm." "So much the better," replied Dumas. "If you know nothing about the subject, you will have no other ideas than those which you will derive from your own observations."

Being at length persuaded to undertake the duty, he had to decide upon the method to be adopted in his endeavour to discharge it. For seventeen years hypotheses and observations with

regard to the disease had been accumulating, and facts and opinions were only too abundant to be made available to any extent in this direction. But among the memoirs which the calamity had called forth, one of the best was that presented to the Academy of Sciences by M. Quatrefages, and a paragraph in it had especially attracted the attention of Pasteur. This had reference to the discovery by some Italian naturalists of microscopical bodies—vibratory corpuscles—in the silkworms and moths, which Lebert affirmed could always be detected in these when diseased, and which Osimo, of Padua, had also perceived in silkworms' eggs. Another Italian, Vittadini, had even proposed the examination of the eggs by means of the microscope, in order to obtain sound ones. The mention of this in the 'Memoir' in question was merely casual, being considered of doubtful importance; but it fixed Pasteur's mind on the necessity for the employment of the microscope—an instrument which had already rendered him such immense service in his experiments on ferments, that its employment again as a means of research possessed a strong fascination for him.

He started on June 6, 1865, for Alais, where the plague raged most disastrously, determined not to return until he had mastered everything of importance connected with it. In a few hours after his arrival he had discovered, and was able to show, the corpuscles in certain worms, and after some days' examination satisfied himself that these living disease-corpuscles were numerous in the chrysalides, while there was not one of the moths but had them in profusion. In the eggs and the worms the germs existed in an imperceptible condition, and the only infallible method of procuring healthy eggs, Pasteur insisted, was by having recourse to moths free from the corpuscles. This method he had proved by experiment to be correct, but critics would not accept his statements, and he pursued his enquiry with that scrupulous care, intelligence, and pertinacity so characteristic of him, returning annually to Alais for several months during five years, to complete his work. The contagious nature of the disease—which was doubted by many, and the manner in which the contagion was conveyed—about which there existed several opinions among those who believed in its communicability, were the first points he determined. To ascertain its contagiousness, as well as the mode of infection, he took some healthy worms free from corpuscles, and fed them with diseased worms, which were pounded and smeared over the mulberry-leaves they ate; and after a certain time the corpuscles, which had already shown themselves in the walls of the intestines, began to appear in the other organs, with those red spots on the heads and the rings of the bodies, which had caused the disease to be named "pébrine" by

the peasants. Digestion in the worms was impeded because of the presence of these organisms in their intestines, and they were generally unhealthy ; while those which spun their cocoons produced chrysalides which were little better than pulp. It was thus shown that the corpuscles, gaining access with the food into the intestinal canal, infested all the body in a few days ; the spots on the skin were only the effect of the disease, being somewhat allied to the eruption of measles in mankind. Another lot of healthy worms fed on untainted mulberry-leaves at the same time, remain perfectly free from the malady. This and similar experiments were repeated, and varied times without number, so as to prove the correctness of the conclusions at which he had arrived, and exactly accounted for what took place in the silkworm establishments. From the malady which attacked the worms at their birth, destroying a whole cultivation, down to the invisible disease that lurked in the cocoon, all was explained. But as in these establishments worms were never directly affected through food purposely soiled by diseased worms, it was asked how they became diseased. Pasteur pointed out that, in a cultivation of silkworms in which there were diseased ones, these were continually fouling the food by their excreta, which the microscope showed to be swarming with these corpuscles. This cause of natural contagion was rendered all the more effective, because the worms, by the weight of their bodies, pressed the excreta against the leaves in crawling over them. By mixing these excretions with water, and painting mulberry-leaves with them, a single leaf off the latter enabled Pasteur to infect as many worms as he liked. Another natural and direct cause of infection was inoculation by the hooks on the feet of the worms, which, in crawling over each other, wounded their skins ; and if these hooks were soiled by infected excretions, or by the corpuscles immediately beneath the skin, infection was certain. Infection at a distance through the medium of the air and the dust it carries, was an equally well-ascertained fact. It was sufficient to sweep the breeding-houses, or shake the hurdles on which the silkworms were reared, to raise the dust of dead worms and corpusculous excretions, which falling over the hurdles of healthy worms, after a time caused the disease to appear among them. When quite healthy worms were placed in a breeding nursery at a considerable distance from unhealthy worms, they, in their turn, became infected. It was observed that in some factories healthy worms were reared, in which the year before nearly all had perished. Pasteur explained this by showing that dust can only act as an infective matter when it is fresh ; corpusculous matter loses its virulence when thoroughly

dried, and a few weeks are sufficient to bring about such a result. The corpuscles contained in the eggs alone caused the transmission of the disease to future generations; and thus it was both contagious and hereditary.

Pasteur had demonstrated that moths free from corpuscles produced eggs also free from them; and also that eggs hatched at a distance from infected eggs produced healthy worms, chrysalides, and moths. It was easy, therefore, to multiply cultivations free from the disease; and to secure the production of silkworms and silk, to guarantee that the eggs were sound, Pasteur recommended that a moth should be crushed up in a little water, which is then examined with a microscope to ascertain if any corpuscles are present.

In his investigations, Pasteur soon recognised that there was another disease at work, no less destructive, though less widely spread, than the so-called *pébrine.* During his experiments, in a cultivation of say a hundred worms, a large number, as many sometimes as twenty, would be picked up daily; these turned black and putrefied within a few hours, being soft, flabby, and hollow; they were free from the *pébrine* spots, and no corpuscles could be found in them, while these organisms were also absent from the chrysalides and the moths of the few worms which were able to spin their cocoons. Pasteur was certain that he had to deal with a distinct disease, and that this was one known to French writers upon silkworms by the name of *flacherie*, or *morts-flats.* The cultivations most seriously affected by the malady came from eggs yielded by moths entirely free from corpuscles. Microscopical examination settled the question. If, at the period of rearing of silkworms, which happens when the mean temperature of the air is high, some mulberry-leaves are crushed with a little water in a mortar, and the liquid allowed to stand for twenty-four hours, it will be found to be teeming with microscopic organisms—some motionless, resembling little rods or spores joined like strings of beads; others, flexible and moving about in a sinuous manner, like the vibrions found in nearly all decomposing organic infusions. The germs of these organisms were on the surface of the leaves before they were pounded, in the water, or on the pestle and mortar. So long as the intestines of the worms were healthy, the germs were either digested or expelled without causing damage; but whenever digestion was impaired, which, with such a ravenous creature, and rapidly growing, might be frequent, then the disease appeared. "Every *ver flat* (flattened instead of round) is one which digests badly, and, conversely, every worm which digests badly is doomed to perish of

flacherie, or to furnish a chrysalis and a moth, the life of which, through the injury produced by organised ferments, is not normally perfected," said Pasteur.

The vitality of the germs of *flacherie* was much greater than those of *pébrine*, which, if dried, perished within a year; whereas the former retained their life for several years. The dust of a silkworm nursery infected by *flacherie* appeared under the microscope largely made up of cysts or spores of vibrios, which would remain latent until wet or damp roused them into activity. Falling on the leaves which serve as food, they are carried into the intestines of the silkworms, grow and multiply there, and cause illness, unless the worms are vigorous, when they may be digested with the food. It is a struggle for life between the silkworms and the vibrios, in which the former are unfrequently victorious.

Accidental *flacherie* could be prevented by hygienic precautions; but when hereditary, *i.e.* developed through diminished vitality of eggs or embryo, Pasteur resorted to the microscope, by which information as to the health of the worms, chrysalides, and moths for egg production, could be obtained.

After the most patient study of this compound disease of silkworms, Pasteur had arrived at such knowledge of its causes, course, and different manifestations, that he could at will produce either form— *pébrine* or *flacherie*, and could so regulate its intensity as to have it appear, experimentally, on a given day, almost at a given hour. But a remedy for the evil was not so readily devised. One form, *flacherie*, was capable of comparatively easy control, by care in feeding, and keeping the surroundings in a proper state.

With the other form—*pébrine*—by far the most serious, preventive measures were not so soon arrived at; but through a series of observations, as simple as they were ingenious, Pasteur concluded that the cause was to be combated in the eggs, and, acting on this view, he ultimately triumphed over the scourge.

The process now most successfully pursued in the silk-growing districts, for avoiding the plague, is described as follows:—

The cocoons are finished, and the appearance of the moths alone is waited for. They arrive, and they pair. Then begins the work of the cultivator, who is careful about the production of the eggs. He separates the couples at the end of the day, laying each female moth by itself on a little linen cloth suspended horizontally. The females lay their eggs. After the laying, the attendant takes each female in turn, and secures her by a pin passed through the wings to a folded corner of the little

cloth, where are grouped some hundreds of eggs which she has laid. The male moth might be pinned to another corner of the cloth, but the examination of this creature is unnecessary, as he does not convey the disease. The female moth, after having been thoroughly dried by free contact with the air, is examined at leisure—as during the winter or autumn. Nothing is easier than to discover whether there are any *pébrine* corpuscles in its dead body. The moth is crushed in a mortar and mixed with a little water; then a drop of this is examined by means of a microscope. If corpuscles be found, the bit of cloth corresponding to the examined moth is known, and burnt with all the eggs it contains.

As has been mentioned, this protective or preservative measure has been found capable of unlimited application, and is universally adopted. In the Basses-Alps, in Ardèche, in Gard, in the Drôme, and in other parts, may be met with everywhere, during the cultivation season, establishments where hundreds of women and young girls are occupied, with a remarkable division of labour and under the strictest supervision of skilful overseers, in pounding the moths, in examining them microscopically, and in sorting and classifying the little cloths upon which the eggs are deposited.

When he had completely fulfilled his mission in 1869, and had restored to France what might have been looked upon as a lost industry,—through the fatigue and intense application to which he had subjected himself,—he was attacked with paralysis of the entire left side of the body, which left him prostrate for some time, and from which he has not yet entirely recovered. His proposed method of combating the silkworm-disease was not accepted without serious doubts and opposition,—for throughout his career he has had to contend continually with determined opponents, who have endeavoured, notwithstanding his lucid demonstrations, to ignore or minimise the grand results he has achieved. In 1869, while still suffering from the severe effects of his unfortunate illness, he, impelled by these doubts and contradictions, had himself carried to Alais, where he struggled, by giving directions from his arm-chair, through a repetition of his experiments, and succeeded in once more proving the certainty and simplicity of his method. The French Government, nevertheless, influenced by his detractors, still hesitated to adopt his process of culture; but the late Emperor interposed, and offered Pasteur the Villa Vicentia, near Trieste, belonging to the Prince Imperial, as a suitable locality for affording a convincing test of the plan, as for ten years the silk-harvest at that place had not sufficed to pay the cost of the eggs. Transported across France and Italy with difficulty, because

of his helplessness, he succeeded in reaching the Imperial Villa, where his skill was rewarded with marvellous success; for in a short time the sale of the cocoons realised a net profit of twenty-six millions of francs. On this result being made known, the Emperor nominated him a Senator of France in July 1870; but before the nomination was gazetted in the official journal, the war with Germany interfered, and Pasteur returned to France to share in the misfortunes and mortifications of his country during that eventful period.

When the war was over, he commenced his studies on the preservation of beer, to which allusion has been already made. The results of these studies were so beneficial that they were universally recognised; and as an instance of the estimation in which they were held beyond France, it may be mentioned that a celebrated scientist of Copenhagen, Jacobsen, had a bust of Pasteur placed in the *salle d'honneur* of his celebrated laboratory. On the conclusion of the report which he made on these studies, Pasteur alluded to the principles upon which, for twenty years, he had pursued his labours—principles the application and advantages of which seemed to him without limit; and, firm in his conviction, he prophetically wrote, "the etiology (cause) of contagious diseases is on the eve of having unexpected light thrown upon it."

Two hundred years before Pasteur had achieved his well-deserved fame, England could boast of three men whose names yet stand pre-eminent in natural science—Newton, Hooke, and Boyle. Boyle was born in the year in which the great Bacon died, and he was the earliest, though perhaps not the most distinguished, of those who practically applied the precepts set forth in the 'Novum Organum' of the Imperial Verulam, and is therefore the patriarch of experimental science, at least in England—a science of which Pasteur is now one of the chief exponents. Boyle's genius as a physicist led him into many notable discoveries and surmises, and one of the latter is most memorable in relation to the subject which now occupies our attention, forecasting, as it did, the revelations effected by Pasteur in the direction indicated by him. "He that thoroughly understands the nature of ferments and fermentation," wrote Boyle, who had devoted much time, amid his multifarious studies, to this question of fermentation, "shall probably be much better able than he that ignores them, to give a fair account of divers phenomena of certain diseases (as well fever as others), which will perhaps be never properly understood without an insight into the doctrine of fermentation."

The mention of contagious diseases recalls sad memories of agricultural misfortunes in the United Kingdom, for nearly half

a century—of desolating plagues among all kinds of stock, for years unchecked and uncontrolled, then encountered with harassing and sterile legislative measures, until property of the value of many millions of pounds was lost, and agriculture was greatly crippled. The admission and the continuation of these maladies amongst our flocks and herds were due, at first, to the little that was known of their history or nature; and in later years, when their communicability was conclusively ascertained, their widespread ravages were unchecked, partly through indifference, and partly through mistaken economical notions.

It was not until the true nature of fermentation was elucidated, thanks to Pasteur, that the cause of communicable disorders in man and beast was satisfactorily and firmly established. The doctrine of a living infecting agent—a *contagium vivum*—in these maladies had certainly been accepted by physicians for a very long time, though it was based only on hypothesis; and even long before then, again, it was imagined, in explanation of the phenomena of contagion, that in transmissible maladies there was given off an imponderable and subtle matter—an *aura contagionis*—which was the active principle operating in their production; this unknown *something* could originate the disorders in healthy bodies into which it had penetrated from those already suffering from its effects. Then the chemical notion of the cause of fermentation, originated mainly by Liebig, was applied to these diseases, which were, and are now, consequently designated *zymotic* (from the Greek word for leaven). Like the ferments, the poisons and disease processes were supposed to be the results of atomic disturbance peculiar to substances in course of molecular change, and capable of communicating themselves to the various constituents of the living body. As with fermentation, so with contagious diseases; the part played by the microscopic organisms, as shown by Pasteur, revolutionised our ideas and revived the doctrine of living contagia. And Chauveau, the eminent director of the Lyons Veterinary School, was the first to demonstrate that the virus of a contagious disease (sheep-pox) was particulate, while putrefaction in wounds was due to atmospheric germs. In 1862, also, Pasteur had announced in his memoir on Spontaneous Generation, and in opposition to the views then entertained, that when urine becomes ammoniacal the alteration is caused by a microscopical fungus, and not by mucus or pus; at a later period he established the fact, that in affections of the bladder ammoniacal urine was always associated with the presence of this fungus, to the development of which borax, he discovered, was antagonistic. The application of this discovery in the treatment of urinary disorders has been most beneficial; as the dangerous fermentation of urine in the

bladder is prevented by injecting a solution of borax, which causes no pain, into that organ. About this time, also, Lister, then a professor of surgery in Scotland, moved thereto by the researches of Pasteur, began his investigations into antiseptic measures in surgical operations, which he has since perfected to such an extent, that "Listerism" in the treatment of wounds is regarded throughout the civilised world as one of the greatest improvements in modern surgery: rendering the healing of formidable injuries more rapid and certain, and enabling the surgeon to boldly and successfully undertake operations which previously he would not have dared to venture upon, or which were in the great majority of cases fatal. Lister's method simply consists in excluding putrefactive and other germs from wounds, as well as from naturally closed cavities of the body which may have to be opened for the cure of disease.

Pasteur was naturally reluctant to enter upon the study of contagious diseases, though, from his success in elucidating the causes of fermentation and the silkworm disease, he hoped to gain, and indeed felt certain of arriving at, an exact knowledge of their origin. "I am neither doctor nor surgeon," he repeated, when urged to undertake researches into the transmissible disorders of men and animals. But at length he was induced to begin, and he selected for his study one of the most widespread and fatal of animal diseases—that which is known to scientists as Anthrax, and to agriculturists and others in this country by various names, some of them local: such as Splenic Apoplexy, Splenic Fever, &c.; and in France as "Charbon," and in Germany as "Milzbrand."

This anthrax destroys wild as well as domesticated animals, herbivorous creatures and rodents being most susceptible, and it prevails, in one or more of its diversified forms, over probably the entire surface of the globe. It sometimes decimates the reindeer herds in the Polar regions, and is only too well known in the tropics and in temperate latitudes. The carefully-tended ruminants of the most highly civilised countries, suffer equally with the wandering herds and flocks of the Tartar Steppes; and the scourge is as much dreaded by the Finns and Lapps, as it is by the Mexicans, the Arabs, the Annamites, or the South African and Australian colonists. It has been carefully described by travellers, veterinary surgeons, and others, as they have observed it in Siberia, Lapland, Russia, Central Asia, and Asia Minor, China, Cochin-China, the East and West Indies, Peru, Paraguay, Brazil, Mexico, North America, Australia, North and South Africa, and Egypt. Europe appears to be specially visited by it, and some parts suffer most seriously from its ravages. The books and papers which have been published

with regard to outbreaks of the disease, and its nature, characteristics, and the damage it inflicts, are innumerable. Countries with extensive marshes, low-lying valleys, or a tenacious subsoil, are those most affected. Thus it happens that there are regions notorious for the prevalence of anthrax—as the marshes of Sologne, Dombes and Bresse, Brie and Beauce, in France; and certain parts of Germany, Hungary, and Poland. It is enzoötic in the half-submerged valleys and the maritime coasts of Catalonia, and also in the Romagna and other marshy districts of Italy; while it is epizoötic, and extends itself to people, in the swampy regions of Esthonia, Livonia, Courland, and, above all, in Siberia, where sometimes, in order to suppress the ravages of the terrible "Jaswa," as it is termed, the aid of the military authorities is called for, and battalions of soldiers are sent to bury or burn the thousands of infected carcasses of animals which have died from it. And elevated countries are not exempted from it, for in the Bavarian Alps, for instance, it exacts an annual tribute of victims.

The antiquity of anthrax is as great as its geographical extension is wide. It was one of the plagues with which the Egyptians were punished in the time of Moses, when there was a breaking forth of *blains* upon man, and upon beast, throughout all the land of Egypt: upon the horses, upon the asses, upon the camels, upon the oxen, and upon the sheep. Virgil, in his 'Georgics,' has depicted its deadliness and its contagiousness with the greatest accuracy, pointing out the dangers of the tainted fleeces of diseased sheep to mankind, as if he were describing the cause of what is now known as the "woolsorters' disease"—anthrax conveyed to man by infected wool, and far from uncommon among people employed in woollen factories in this country. It frequently occurs in the histories of the Early and Middle Ages, as a devastating pestilence among animals, and through them as a plague of mankind. Our oldest Anglo-Saxon manuscripts contain many fantastic recipes, charms, and incantations, for the prevention or cure of the "blacon blegene" (black blain), and the relief of the "elf-shot" creatures. From these up to our own times, anthrax has attracted more and more attention; even in this century, it has spread in some of its outbreaks over the whole of Europe, from Siberia to France.

The losses inflicted by anthrax are appalling. Some idea of their extent may be derived from the fact, that in one district of France alone, Beauce, it kills about 178,000 sheep, which (at only thirty francs a head) are worth 5,340,000 francs, or 213,600*l.* In 1842, when sheep were much less valuable than now, the loss in the same district was estimated at 7,080,000

francs. The disease also prevails among sheep in Brie, Champagne, Berri, Poitou, Auvergne, Dauphiné, and Bourgogne. In the arrondissement of Chartres, 17,800 perish from it every year; in Beauce, 20 per cent. of the sheep die, a loss of seven or eight millions of francs annually. It is estimated that sheep to the value of twenty millions of francs are lost annually in France. Cattle, horses, and other creatures, suffer also severely. In Russia the losses are enormous, especially among the horses and cattle. In 1837, in one district alone, 1900 died of anthrax; and in 1857, for the Russian Empire, it was reported that 100,000 horses had perished. In 1860, 13,104 cattle, out of 18,883 attacked with the "Jaswa," succumbed; and from the official report for 1864, it appears that in the five governments of St. Petersburg, Novgorod, Olonetz, Tver, and Jaroslav, 10,000 animals died, most of them horses, few cattle, and still fewer sheep; while 1000 persons were infected and perished. From the 15th of January to the 27th of March, 1865, 47,000 cattle, 2543 horses, and 57,844 other domesticated animals, were lost in the governments of Minsk, Vitepsk, and Mohilev; and in the government of Tobolsk, in June and July, 1874, there perished from the "Siberian plague" (as anthrax is sometimes designated) 4735 horses, 516 cattle, 1030 sheep, 52 pigs, 15 goats, and 106 human beings. In other European countries it is very prevalent and deadly, and in our own islands it causes heavy losses at times. In India it is witnessed in all animals, and as "Loodiana disease" it is well known as a fatal scourge among military horses. In South Africa, as "Horse-sickness," it is most destructive, particularly in low-lying damp regions, at a certain season of the year, when it kills nearly all the horses after only a few hours' illness.

The real cause of anthrax was, until recently, involved in the greatest obscurity, and many influences were invoked to account for its appearance. But it may be noted that several Continental veterinarians, as Numann, Marchant, Gerlach, Plasse, and Delafond, were of opinion that it was of cryptogamic origin—the minute fungi finding entrance into the body with damaged grain or forage. The contagious nature of the disease had been known from the remotest antiquity.

In 1844, Delafond, then a teacher at the Alfort Veterinary School, was sent by the French Government to investigate the disease as it appears among sheep—known as the *maladie du sang*—in the districts where it was causing the heaviest losses. On examining the blood of diseased sheep microscopically, he observed peculiar rod-shaped bodies, but did not attach any importance to them. This fact has been lost sight of by nearly all recent writers on the malady. Pollender, a

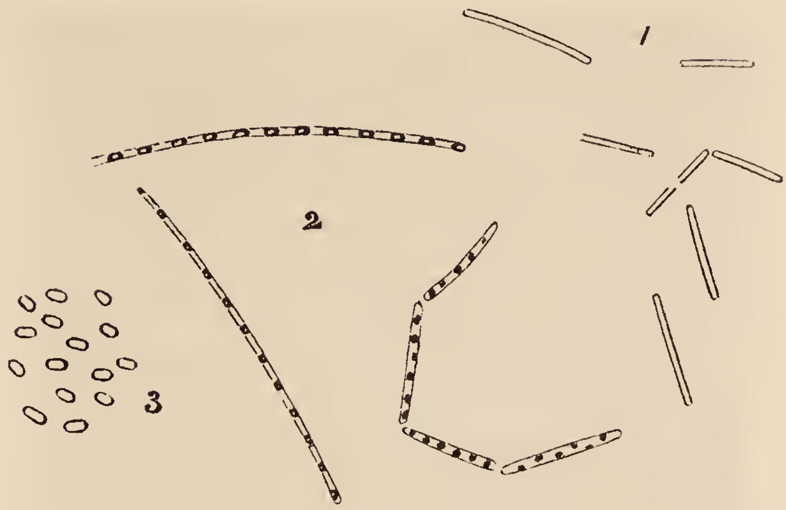
German, observed these bodies in 1849, Davaine and Rayer in 1850; and Brauell, of the Dorpat Veterinary School, in 1856, also noticed them. But these likewise did not realise the significance of their presence in the blood of anthrax-stricken animals.

It was not until 1863, that Davaine, having had his attention once more drawn to these organisms in the blood, by Pasteur's discovery of the butyric ferment, was led to examine whether they, if introduced into the blood of an animal, would also play the part of a ferment; as, if so, this would explain the rapid infection of the blood in an animal which had received into its veins, accidentally or experimentally, a certain quantity of this ferment. Inoculating sheep and rabbits with blood taken from a diseased sheep a few hours after death, he constantly found the organisms, which he named *Bacteria;* though when inoculated with blood from the infected animal before these rods were visible, and while it yet appeared in health, no effect was produced. "In the present state of science," wrote Davaine at this time, "no one would think about going beyond these corpuscles to seek for the agent of contagion. This agent is visible, palpable; it is an organised being endowed with life, which is developed and propagated in the same manner as other living things. By its presence and its rapid multiplication in the blood, it undoubtedly produces modifications in that fluid, after the manner of ferments, which speedily destroy the infected animal." This was the first direct evidence that a contagious disease is produced by a microscopic organism, and the discovery converted into a fact what was before only a hypothesis.

Davaine's announcement was not received without doubt and hesitation by some, and two experimentalists—MM. Jaillard and Deplat—were unsuccessful in arriving at the same results by experiment, and consequently sought to refute his conclusion. Paul Bert also opposed his view, and corroborated that of Jaillard and Deplat. "I can," he said, "destroy the bacteria in a drop of blood by means of compressed oxygen, inoculate with it, and reproduce the disease and cause death without any appearance of bacteria. Therefore they are neither the cause nor the necessary effect of splenic fever. It is due to a virus." And so the controversy went on.

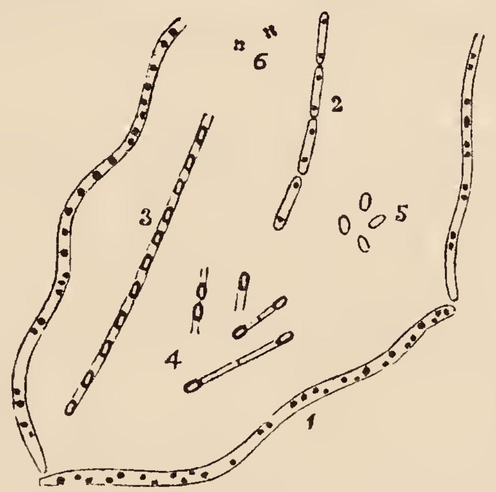
At this time, a young physician in Germany, Dr. Koch, began to study the anthrax *bacilli* or *bacteria*, as they have been indifferently termed; though Cohn, another German investigator, was also at work in defining their morphology. Koch raced the life-history of the *Bacillus anthracis*, as it is now designated, and showed how widely it was distributed throughout the blood and organs, and especially the spleen of animals which

Fig. 5.—*Bacillus Anthracis.*



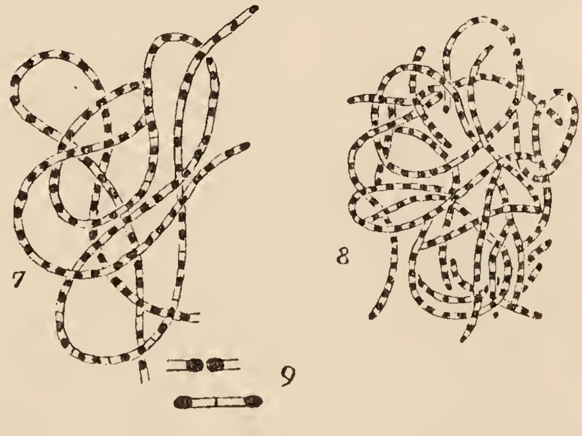
1. Rods of *Bacillus anthracis* as seen in the blood. 2. Portions of rods under cultivation. 3. Groups of spores. 1 and 2 magnified about 500 diameters. 3 about 700 diameters.

Fig. 6.—*Stages of Bacillus Anthracis under Cultivation.*



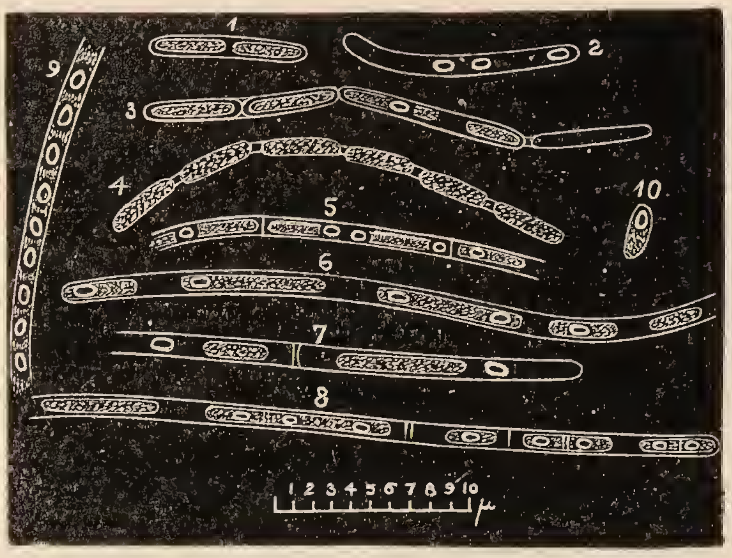
1. Wavy filament, showing commencing lateral formation of spores, as granular dots. 2. Another part of same cultivation, filament dividing, each segment having a terminal dot. 3. Filament in which spores have formed at nearly regular intervals. 4. Portions of filament with spores after division. 5. Isolated spores. 6. Sporules formed by division of spores.

Fig. 7.—*Further Stages of Bacillus Anthracis under Cultivation.*



7. Part of convoluted filaments in which spores have formed, and division is commencing in parts, from cultivation of *Bacillus anthracis.* 8. Similar process in another *Bacillus* not connected with anthrax. 9. Portions of filament from 7 more highly magnified.

Fig. 8.—*Bacillus Anthracis, magnified* 1500 *diameters.*



1. A short rod (8 μ long) in which the protoplasmic contents have divided previous to fission of the rod. 2. A clear rod containing only spores. 3. A jointed filament composed of three segments, in one of which fission is commencing, in another a spore has formed, whilst another is empty. 4. Part of filament composed of short homogeneous rods united by a narrower sheath. 5, 6, 7. Parts of long filaments showing various arrangements of the protoplasm and spores. 8. From another filament in which spore formation is proceeding more regularly. Commencing fission is seen at some parts of the filament. 9. Part of filament completely filled with regularly arranged spores. 10. From another cultivation, showing sporule germinating into a short rod.

had died from anthrax; after many attempts he succeeded in growing, or "cultivating," the fungus artificially, and watching its development under the microscope: noting that the rods, like those of the butyric ferment, multiply by division, grow into long straight or twisted filaments, in which, in the course of time, if air is allowed free access, very minute oval spores are seen; then the filaments swell and burst, and the spores are set free. These spores, if introduced into the blood of an animal, or cultivated artificially, grow again into the characteristic bacilli, which eventually become the long filaments that form spores. Such is the life-cycle of this deadly microbe, which multiplies so rapidly that the spores have been seen to germinate in three or four hours; and rodents—such as rabbits, guinea-pigs, or mice—when inoculated with the blood of an animal dead of anthrax, or with the bacilli or spores of an artificial culture, perish usually in from twenty-four to forty-eight hours. The blood, which is dark-coloured, always contains these bacilli, as does the spleen, which is greatly enlarged, sometimes immensely so, and soft, like a bag of water; the urine, as well as the exudations thrown out during the life of the animals, likewise contains these bodies.

Pasteur possibly did not know of Koch's admirable researches, which were published in 1876; neither, probably, did the other disputants in France, for it was in that year that Bert gave an adverse opinion to that of Davaine. However this may be, Pasteur determined to settle the question as to whether

Davaine or his opponents were in the right: and he adopted in his investigation the methods which had led him to such splendid results in his previous studies. He resolved to isolate the organism from infected blood, cultivate it, as Koch had done, in artificial media, and then to test its action upon animals. In this investigation he was assisted by M. Joubert, as, owing to his paralysis, he could not carry it on by himself.

In 1877, he reported to the Academy of Sciences that the bacilli were the only agents in producing anthrax. A little drop of splenic-fever blood sown in urine or in yeast-water, previously "sterilised" (rendered imputrescible by contact with air free from germs), in a few hours produces myriads of them; a drop of this first cultivation sown in a similar manner and with the same precautions, proves as fertile; and this process being repeated for ten or twenty times, it may be taken for certain that all the substances which the first drop of blood might have carried with it, have been completely got rid of. Yet Pasteur found that if a very small quantity of the final cultivation were injected beneath the skin of a rabbit or a sheep, it killed them in two or three days with all the symptoms of the natural disease. In order to satisfy himself and others that there was nothing else than the bacillus at work in producing this effect, he had his cultivation-fluids kept at an absolutely uniform temperature, which allowed the organisms to be deposited at the bottom of the tubes. When he inoculated with the clear upper fluid, or the deposit—the bacilli—he found that the latter alone caused disease and death.

But it was necessary to explain the discrepancy between the results obtained by himself and Davaine, and those of Jaillard and Deplat. These latter had speedily killed rabbits with blood from a cow that had died of splenic fever, and their blood was also so virulent as to kill others, and yet no bacilli could be found in it; while Bert maintained that he had destroyed the bacilli by compressed oxygen, and still the blood retained its virulence. It might be surmised that there were two kinds of destructive agents operating in this blood, and Pasteur had the good fortune to demonstrate that this was actually the case. Some years previously he had ascertained that in none of the tissues or fluids of the body, except in the intestinal canal, are there any germs; that canal alone contains them, as they readily find access to it along with the food and water, and its temperature is favourable to their development. In the commencing portions of the intestine, in which there is air, ærobic microbes can exist; but toward the terminal divisions oxygen is absent, and only anærobic microbes can live. During life the latter can do no harm, as the vitality of the

lining membrane of the intestines prevents their passage through it; but after death this resistance ceases, and the anærobic bodies then begin to exercise their decomposing influence, and carry on the process of putrefaction—penetrating into the tissues and the blood as soon as that has lost its oxygen. In splenic fever, the blood is deprived of much of its oxygen during the course of the disease, so that, after death, decomposition is very rapid. The germs or vibrions which most readily and rapidly pass to the blood after death, are the septic or putrefactive, which cause the evolution of most fetid gases, by their action upon nitrogenous and superfluous matters. So that after twelve or fifteen hours, the blood of a diseased animal, which, at the time of its death, and for a few hours after, contained only the bacillus of anthrax, has now, in addition, the septic vibrio. The two are antagonistic, inasmuch as the anthrax bacillus, being ærobic, soon perishes in blood destitute of oxygen; while the septic, being anærobic, is, on the contrary, in the most favourable condition for development, and speedily invades all the fluids and solids of the body. From this circumstance, it happens that if a drop of blood is taken from the body of an animal that has just died from anthrax fever, and is inoculated into a healthy animal before it has had time to putrefy, anthrax fever only will result. But if, on the contrary, the operation is performed after a longer time, when putrefaction has taken place in the blood, then inoculation will produce at one and the same time splenic fever and septicæmia, or putridity of the blood: both being developed simultaneously, or one before the other, usually septicæmia—the septic vibrio causing death before the anthrax bacillus has had time to multiply in sufficient numbers to bring about such a result. This affords an explanation of the different conclusions arrived at by the experimentalists. Jaillard and Deplat had obtained blood which was more or less putrid, and therefore contained the vibrios of putrefaction mixed with the organisms of splenic fever; it was therefore septicæmia, and not anthrax which had killed the rabbits; and in examining their blood the septic vibrios had escaped notice, while no bacilli were observed. So that when Davaine asserted that these two opponents had not used pure anthrax blood, he was right, though he could not give his reasons. Bert's apparently contradictory result was also accounted for in the same way; he had employed blood which was more or less putrid, and as the spores of the septic vibrio perfectly resist the influence of compressed oxygen, these had not been killed, while the filaments of bacilli and septic vibrios had perished. The inoculated animals died from these spores. To prove that this was so, Pasteur resorted to the method of

successive cultivation of them in an artificial medium, and in as perfect a vacuum as possible, or in contact with carbonic acid gas, but no air, as this kills them—conditions the opposite of those in which the anthrax spores develop. From blood containing these two organisms of anthrax fever and putrid fever (septicæmia), he could produce either disease experimentally, at will, according as he cultivated the germs with or without air; or by inoculating with blood from an animal just dead from the former malady, or obtained from it twenty-four hours after death.

These discoveries clearly established the correlation between the phenomena of fermentation and those of contagion. There is no longer anything mysterious about the latter. It is simply the breeding of a living organism, or "element," in a living body, and at the expense of that body; in the same way as the organism of fermentation lives and multiplies in, and at the expense of, the dead organic matters into which it has found its way. Just as the contagious itch, or "mange," is produced on the surface of the skin by a very small insect or mite (the *Acarus scabei*); and "ringworm," also contagious and situated on the skin, is due to a microscopic vegetable fungus (the *Tricophyton tonsurans*); so are what we may term the "internal contagious disorders," some of them comparatively mild, as foot-and-mouth disease; and others most fatal—as anthrax, cattle-plague, and sheep-pox—due solely to living germs which prey upon the bodies of the animals they invade, and for whose fluids and tissues they each have a special predilection. I shall recur to this interesting feature presently. In the meantime, it may be stated that the question as to whether contagious diseases ever arise spontaneously is settled in the negative: spontaneous contagious disease has no more foundation than the spontaneous generation of organised bodies; and I need not discuss the primary origin of these diseases or the germs which occasion them, any more than I need do the origin of potatoes, turnips, dogs, or horses, when studying the attributes, derivation, or natural history of these or any other vegetable or animal body.

When studying splenic fever, Pasteur was struck by the fact that the microbe which causes it is under a particular influence which prevents it developing everywhere, and limits it to certain media which, one would think, do not differ from other media in any appreciable manner. Thus in the ox, sheep, rabbit, guinea-pig, and some other creatures, it is readily inoculable and terribly fatal; but the dog and pig have to be inoculated several times before they are infected, and even not then with certainty; while fowls are proof against the disease.

Why the latter should enjoy immunity when a large quantity of anthrax blood was introduced into its body, and an ox would succumb to a minute quantity, was found to be owing to the fact that the microbe will not grow when subjected to a temperature of 112° Fahr. The temperature of birds being from 105° to 107°, Pasteur was of opinion that their blood was too warm for the germination of the microbe, which could take away the oxygen from the blood-globules with much more difficulty than from the air in artificial cultivations ; he, therefore, lowered the heat of the fowls, having previously inoculated them, by immersing their legs in water at 77°, when their temperature went down to 98° or 100°,and in twenty-four hours they were dead, their blood swarming with bacilli. To demonstrate still more forcibly that temperature was the cause of exemption in fowls, a hen, with its body heat reduced as in the other instance, after having been inoculated with anthrax blood, became feverish, and when the fever was at its height it was taken out of the water, wrapped in cotton wool, and placed in an oven at a temperature of 95° ; it gradually rallied, and in a few hours was well. Fowls killed after being rescued in this way, showed no trace of Bacilli in their bodies. I have not heard that this method of saving fowls from the effects of anthrax inoculation, by raising their temperature, has been tried in the case of people suffering from "Wool-sorters' disease," or in that of animals affected with anthrax ; but in the treatment of human typhoid fever, which is doubtless due to a microbe, good results have been reported from cooling the body of the patient by repeated baths in cold water. This may be due to the diminished temperature more or less arresting the fermentation caused by the disease germs.

In my work on 'Veterinary Sanitary Science and Police' (vol. ii., p. 208), I have described a disease affecting poultry, which, from some of its symptoms, and from its sometimes occurring in this century coincidently with human cholera, has received the name of "Fowl Cholera." In my 'History of Animal Plagues,' however, I have shown that it has no relationship, in point of time, with the terribly fatal scourge of mankind ; and in the first-mentioned work, I have demonstrated that it differs widely from it in its pathology. It has been known from a very early period in Europe, and it has appeared in India and on the American Continent—everywhere manifesting itself as a very rapidly destructive disorder. Since 1798, it has been studied and described in Italy, Germany, and France by veterinarians and others. Delafond, who first saw the anthrax microbe, believed it to be that disease ; in 1873, a veterinary surgeon of Alsace, Moritz, discovered a special microbe in the blood of diseased fowls ; and in 1878, Professor Perroncito, of the Turin Veterinary

School, described this organism; while Professor Toussaint, of the Toulouse Veterinary School, also recognised it, and sent Pasteur the head of a bird that had succumbed to the malady. It had, during this century at least, been considered a most virulent disorder, readily communicable to healthy fowls by cohabitation, by inoculation, and by feeding them with food or water tainted by diseased birds.

Pasteur cultivated the germs in fowl-broth, in which it multiplies in a wonderful manner, as in a few hours the clearest decoction becomes turbid, and is found to be swarming with extremely small organisms, constricted in the middle, and motionless; but in a few days they change into a lot of isolated specks so minute, that the fluid, from being almost milky white, again becomes almost as clear as it was at first. The organism was found to belong to the kind called "micrococci"—in which class Pasteur predicted the microbes of the viruses yet unknown would probably be found. He observed that yeast-water, in which so many diverse microscopic organisms find suitable nourishment, was quite unsuitable for the growth of this microbe—indeed, it perished in it; and that the fowl-broth was the only medium in which it really flourished. The thousandth part of a drop of this mixture inoculated in a fowl would cause death; but in guinea-pigs only a small abscess appeared at the point of inoculation, and this opening spontaneously, a little pus, teeming with microbes, escapes. The virulence of this pus is extreme, and if fowls are inoculated with it they will perish quickly; but the guinea-pig suffers no disturbance in health. Rabbits appear to be as susceptible almost as fowls; so that if the pus of the abscess were smeared over the food of fowls and rabbits associating with the guinea-pig, these would die, but the latter would remain as well as ever. The excrement contains the organisms in great abundance, and it is chiefly through this that the disease is propagated in poultry yards.

Repeated cultivations do not diminish the activity or potency of the organism of fowl-cholera, which is ærobic, and cultivable in air or aerated fluids; unlike that of anthrax, however, which, when excluded from the air, in a few days disappears or is reduced to fine amorphous granulations, this may be kept for years without air, and still remain active.

In experimenting thus with anthrax virus and that of fowl-cholera, Pasteur was on the eve of his greatest discovery—the attenuation or enfeebling of the virus of contagious diseases, by which the bane could be made to serve as its antidote in the living body. The successive steps by which he arrived at an intimate knowledge of the real agents at work in the production of these diseases, will be observed to be closely related to each

other, as each was the rigorous verification, by conclusive experiment, of a preconceived idea, upon which he always worked. "Nothing can be done," he asserted, "without preconceived ideas; only there must be the wisdom not to accept their deductions beyond what experiment confirms. Preconceived ideas, subjected to the severe control of experimentation, are the vivifying flame of scientific observation, while fixed ideas are its danger."

The fact that certain contagious diseases, as a rule, affect an animal or a man only once in a lifetime, has been known from time immemorial; and this fact of non-recurrence has been acted upon in the case of some of these disorders, in producing them purposely in man and beast, in a milder form if possible, or, at any rate, under more favourable conditions, than in the ordinary accidental or natural manner. Eastern people — the Chinese, for example — looking upon an attack of small-pox as inevitable, and knowing the great mortality or disfigurement caused by the disease when caught through infection, have resorted to inoculation in order to induce a less serious and more tractable form at a convenient time. With the same object, for very many years, inoculation has been practised for sheep-pox in some of those countries—as Germany and France—where it always prevails to a more or less considerable extent. In the Russian Steppes, also, for a long time experiments were made to test the advantages of inoculating cattle for Rinderpest; and in recent years the same measure has been tried for "distemper" in dogs. In some outbreaks of Foot-and-mouth disease, cattle-owners have often produced this troublesome affection in their yet healthy animals, in order to prevent it lingering among them for a long time, and so to get rid promptly of what threatened to be an inevitable and more harassing visitation. And protective or preventive inoculation for the Lung-plague of cattle, introduced by Dr. Willems, of Hasselt, some thirty or forty years ago, has been very much resorted to in countries where that insidious and deadly pest prevails. But in all these instances the inoculation has been effected with the natural, or what we might designate the "crude" virus; and therefore the danger was great that the diseases might at times be induced in as great virulency as when they appeared in the ordinary way. This has been the case especially with Cattle-plague inoculation, which, because of the unsatisfactory results, has now been abandoned; while the benefits to be derived from Sheep-pox inoculations are perhaps more than counterbalanced by the disadvantages which attend them.

The introduction of vaccination as a protection against small-pox, by Dr. Jenner, has proved an immense success, but hitherto

it has been an unique instance of one form of disease proving an antidote to another form. Cow-pox is not the same disease as small-pox, and by no known means can it be transformed into the latter; neither is cow-pox human small-pox changed in almost every characteristic feature by transmission to the cow. Physicians and others have made the strange blunder of considering the two diseases as dependent on the same virus, notwithstanding the most obvious reasons to the contrary; and this blunder is repeated in medical books. But it is a patent fact, nevertheless, that cow-pox virus, when inoculated in man, protects him from small-pox, which the pox of no other animal will do, that I am aware of, except it be that of the camel and horse. The microbes of cow-pox find a congenial soil in mankind, multiply there, and make that soil unsuitable for the microbes of small-pox.

Why one attack of a contagious disease should render the body of a creature proof against another, for a more or less extended period, or even for the entire duration of life, is not at present known. Various hypotheses have been presented at different times to account for it, but that which has perhaps received most favour is the one that explains the immunity, by suggesting that something has been removed from the blood in the first attack which is necessary for the growth and multiplication of the disease-producing germs, and which is only slowly, or never, reproduced. In the words of Professor Tyndall, "When a tree or a bundle of wheat or barley-straw is burnt, a certain amount of mineral matter remains in the ashes, extremely small in comparison with the bulk of the tree or of the straw, but absolutely essential to its growth. In a soil lacking, or exhausted of, the necessary mineral constituents, the tree lives, the crop cannot grow. Now contagia are living things, which demand certain elements of life just as inexorably as trees, or wheat, or barley; and it is not difficult to see that a crop of a given parasite may so far use up a constituent existing in small quantities in the body, but essential to the growth of the parasite, as to render the body unfit for the production of a second crop. The soil is exhausted, and, until the lost constituent is restored, the body is protected from any further attack of the same disorder. . . . To exhaust a soil, however, a parasite less vigorous and destructive than the really virulent one may suffice, and if, after having by means of a feebler organism exhausted the soil without fatal results, the most highly virulent parasite be introduced into the system, it will prove powerless." Another theory, highly favoured by some eminent authorities, is to the effect that the microbes, during their multiplication in the body, cause the production

of some substance which is detrimental to a second invasion of them. This theory would seem to have good foundation in what is known to occur in fermentation. The poisonous substance may remain persistently in the blood and tissues, in which case immunity will be permanently established; or it may be removed in the course of time, and the body become again susceptible, and then the protection is only temporary. Both theories are perhaps equally acceptable, in explanation of what occurs.

Vaccination, as a defence from small-pox, seems to have led Pasteur to ask of himself why, if contagious maladies do not recur, there should not be found for each of them a different disease, but resembling them, which acting upon them as cow-pox does upon small-pox, would prove preventives of them? And what has been called a "chance occurrence" appears to have thrown open the way to enable him to give a reply to the question. Having shown that the microbe of fowl-cholera could be cultivated in an artificial medium for hundreds of times, without its virulence being diminished in the slightest degree, he found that this virulence could only be assured when no great lapse of time had occurred between the successive cultivations—the second culture being sown twenty-four hours after the first, the third twenty-four after the second, and so on to the hundredth or more. When several days, weeks, or even months, were allowed to elapse between the cultures, a progressive weakening in power of the microbe was apparent. So that if fowls are inoculated with the successive cultivations made at short intervals, and die in the course of one or two days, those which are inoculated with that which has been made at an interval of some weeks or months, will suffer much less and recover. But the most startling phenomenon in this respect is, that after recovering, should they be re-inoculated with the most active virus which would kill its hundred per cent. of those inoculated with it in a few hours, they will not die, perhaps even show no signs of illness: they are protected by inoculation with the delayed or attenuated cultivation.

The discovery of this power in the weakened microbe of a contagious disease, to protect an animal from the lethal action of the same kind of microbe when in full virulency, must be held to constitute the most important of Pasteur's services to medical science and to mankind; inasmuch as its effects may be of the greatest magnitude, and far-reaching, when made applicable to the many contagious disorders—so deadly and so little under the control of man—which affect our own species and the lower animals. The destructive microbe is transformed in the laboratory, by the skill of man, and at will, into a benign, life-

preserving agent. In the case of the fowl-cholera germ, the attenuating influence in operation was merely the oxygen of the air; and the proof of this is afforded by cultivating the germ in a tube having only a little air. If the tube be hermetically sealed, the microbe soon consumes the oxygen in the air and in the culture-fluid, and retains its potency for evil unimpaired, for months, or even for years. The reason why the oxygen of the air has no influence upon it during the twenty-four hours' cultivation, is explained by this being required for its nourishment and reproduction; after that time, the air gradually modifies or weakens it, until at last its power is annihilated, though the organism still lives. The experimentalist can in this way reduce, as he wills it, a virus to any degree of virulency. Pasteur could inoculate fowls with a cultivation too feeble to protect from the deadly action of the crude virus, but effectual in insuring them against a stronger cultivation. A second cultivation would nullify the action of a third, and so on until the most deadly virus was rendered inert in the bodies of the fowls. The whole secret of this protective inoculation consists in knowing at what moment a certain degree of virus attenuation is a guarantee of safety against the full-power virus.

Trials of the attenuated virus of fowl-cholera were made by veterinary surgeons and agriculturists, and with perfect success in every instance. It was found necessary to inoculate each fowl twice—at first with a very weak virus, yet sufficiently strong to prevent dangerous consequences from a stronger; then in ten or fifteen days with a less attenuated one. The preparation of these cultivations—or "vaccines," as Pasteur designates them, in honour of Jenner, as it is obvious they have nothing to do with cows in this and some other animal diseases—demands much care in manipulation and experimentation, in order to arrive at the proper degree of harmlesness and potency. Species, and even breeds, and likewise individual peculiarities, have to be tested as to receptivity, before the method can be generally applied.

When Pasteur laid before the Academy of Sciences the details of his new discovery, the importance of its bearings on medicine—human and veterinary—as well as on medical doctrines, was fully recognised by those who were competent to form an opinion, and by no one more enthusiastically than the eminent veterinarian, M. Bouley, director of the French Veterinary Schools, and whose death last November, while President of the Academy of Sciences, Medicine and Agriculture must deplore. "This is but a beginning," he exclaimed; "a new doctrine opens itself in medicine, and this doctrine appears to me powerful and luminous. A great future is preparing; I wait for it

with the confidence of a believer, and with the zeal of an enthusiast."

Having succeeded so remarkably with the plague of fowls, Pasteur was naturally desirous of extending his protective inoculation to other diseases. Anthrax was at that time being investigated in his laboratory; but as nothing was known as to whether one attack of the disease gave exemption from another, no idea could be formed whether inoculation would be successful. People who are accidentally infected from diseased animals rarely recover, and when they do, a second infection is not likely to be observed; sheep attacked with it generally all perish; but bovines are more resisting, and recoveries are not unfrequent. An opportunity occurred in 1879, in a veterinary surgeon of the Jura bringing forward a cure for splenic fever, when Pasteur was deputed by the Minister of Agriculture to report upon its value. A lot of cows were inoculated with crude virus, and one-half of the number were treated according to the veterinary surgeon's method, the other half receiving no treatment. The pretended remedy was a failure, but though many cattle died from anthrax, others survived, after being very ill. When they had quite recovered, they were re-inoculated with large quantities of the virus, but no symptom whatever of disease resulted—even when this was done after more than a year had elapsed. The question of immunity from recurrence was therefore settled, and the object now was to find a method of attenuating the virus of this malady, as he had that of fowl-cholera.

But Pasteur and his able assistants, Chamberland and Roux, met with a serious difficulty at starting. The microbe of anthrax differs from that of the poultry disease, in reproducing itself by spores, as well as by fission, the latter alone being the mode of generation in the last-named malady; while the spores may be exposed to the air for years without losing their virulence, and ready to develop into the characteristic rods, or bacilli, whenever transferred to a suitable medium. The virulence of the spore is protected from injury by its impervious covering or shell. The problem therefore was, supposing the filaments or bacilli to be analogous to those of fowl-cholera, and cultivable as a protective agent, to determine the conditions which would prevent the formation of spores. At length, after many experiments and anxious consideration, success was secured. It was found that at a temperature of 111° to 114° the anthrax microbe could not be cultivated, but that this could easily be done at 108° or 109°, at which it produced no spores. At that temperature, therefore, and in contact with pure air, cultivation of bacilli destitute of spores can be effected. But in a few weeks the crop

perishes, for when sown into a fresh medium there is no result; though in the preceding days life is still present in them. When the cultivation is exposed to air and warmth for two, four, six, or eight days, and is then inoculated in animals, its activity is found to be modified in proportion to the duration of exposure; it presents, in fact, a succession of gradually attenuated viruses, as in fowl-cholera, each constituting a foil for the uncultivated microbe.

The announcement of a method of protection from the most serious animal disease in France, was made on February 28th, 1881, to the Academy of Sciences, where it was received with great acclamation, and immediately afterwards the President of the Society of Agriculture of Melun, on behalf of the Society, invited Pasteur to give a public demonstration of his method of conferring immunity from splenic fever; this invitation he accepted, and sixty sheep were placed at his disposal. It was arranged that ten of these were to be left untouched; twenty-five were to receive an inoculation of very attenuated virus, and in twelve to fifteen days another inoculation of stronger cultivated virus. Some days afterwards these twenty-five sheep, with the other twenty-five which had not been protected, were to be inoculated with the crude mortiferous virus of anthrax.

Ten cows were also given for experiment—six to be inoculated, four not; the ten were afterwards, the same day as the fifty sheep, to be inoculated with uncultivated virus. Pasteur prognosticated that the twenty-five sheep which had not been inoculated would die, and the other twenty-five, protected ones, would survive; also, that the six inoculated cows would resist the poison, while the four which had not been inoculated, even if they did not die, would at least be extremely ill. This prediction startled many of his colleagues at the Academy of Sciences, and some of them designated it a scientific imprudence, which his laboratory experiments scarcely warranted. Pasteur, however, had perfect confidence in the result, as had also his friend and warm admirer, M. Bouley.

On May the 5th, the experiment was commenced on a farm belonging to a veterinary surgeon, M. Rossignol, who was Secretary-General of the Melun Agricultural Society. At the desire of the Society, a goat was substituted for one of the twenty-five sheep of the first lot. Twenty-four sheep, the goat, and six cows were each inoculated with five drops of cultivated virus, this being placed beneath the skin by means of a hypodermic syringe. Twelve days afterwards, these animals were reinoculated with a stronger cultivated virus; and on May the 31st, all of the cattle and sheep were inoculated with very virulent uncultivated virus. A great number of veterinary surgeons, medical and scientific

men were present, and one of the former, in order to render the trial more comparative, had an animal which had been inoculated with the cultivated virus, operated upon with the deadly material alternately with one which had not been protected. Pasteur then arranged a meeting for June the 2nd, which allowed an interval of forty-eight hours after the test inoculation. On that day more than two hundred persons — prefects, senators, counsellors, doctors, veterinary surgeons, and farmers—assembled at Melun, and it is reported that they could not repress a shout of admiration. Of the twenty-five sheep which had not been protectively inoculated, twenty-one were dead, two were dying, and the others were certain to die that evening; the goat also was dead. The non-protected cows were all suffering from intense fever, and were so weak that they could not eat, while there were great swellings where they had been inoculated with the crude virus. The protected sheep were healthy and lively, while the cows did not exhibit the slightest disturbance in health, and there was no tumour at the seat of inoculation. The most incredulous were convinced of the value and reality of the experiment, and were unable to conceal their astonishment at the victory Pasteur had gained over such a deadly disease. A veterinary surgeon who had been the most sceptical, now became such a convert that he had himself inoculated with the two cultivations, without any other accident than a slight fever; and it required all the efforts of his family to induce him to desist from inoculating himself with the unattenuated virus.

On the result of this experiment being made known through the medium of the public press, Pasteur was overwhelmed with applications for inoculating material, or "vaccine," from breeders of sheep and cattle, as well as from Agricultural Societies; so that he was compelled to institute a small manufactory in the Rue Vauquelin, Paris, for its preparation. At the end of 1881, 33,946 animals had been protectively inoculated, of which 32,550 were sheep, 1254 oxen, and 142 horses; in 1882 the number amounted to 399,102, which included 47,000 oxen, and 2000 horses; and in 1883, 100,000 animals were added to the total of the previous year. In the flocks where half had been protected and the other half had not—all herding together—the deaths from anthrax in 1881 were ten times less among the former than among the latter, being 1 in 740, against 1 in 78; in cows and oxen it was 14 times less, being 1 in 1,254 against 1 in 88. In 1883 it was proved that the protection conferred by the inoculation of cultivated virus generally lasted longer than a year; but it has been found prudent to recommend inoculation every year, performing

the operation at a period before anthrax is usually developed—in March and April; for it is recognised that such inoculation is powerless against the disease in a state of incubation—this is, when the poison is developing in the system.

If we consider that France was estimated to lose, annually, from this disease alone, animals valued at twenty millions of francs (800,000*l.*), we need not wonder at the anxiety of agriculturists to avail themselves of any measure which offered a prospect of diminishing such a serious drain upon their resources—not to mention the risk they and their labourers incurred from such a dangerous contagion, as well as the trouble of burial of carcasses, and other disadvantages which cannot be directly estimated.

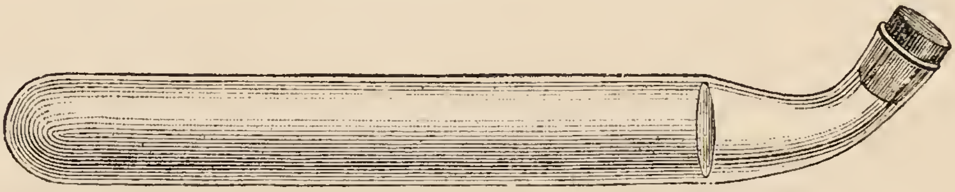
A few words may now be employed in describing, in outline, the method of enfeebling or attenuating the virus of this and some other virulent diseases, in order to procure protective material, or "vaccines," for inoculation, as well as in sketching the manner of performing inoculation.

The crude virus of the disease is first sown, in minute quantity, in what is termed cultivation liquid. But in order to exclude all other germs, except those which are to be cultivated, the greatest care is necessary in order to completely "sterilise" this liquid, previous to the sowing; and everything relating to the operation — cotton-wool, filters, culture-tubes, flasks, and all implements—has to be rendered thoroughly free from germs, or sterile, by exposure to a high temperature, such as will certainly destroy all organisms (298° to 334° Fahr.). The seed-bed, or cultivation liquid itself, which is some simple infusion or decoction of suitable organic matter, is boiled for an hour at a time on two consecutive days, in glass vessels with a narrow neck, which is then plugged with baked cotton-wool; this wool intercepts the entrance of germs by filtering the air, while allowing the latter to pass through. And to make certain that all spores which may have been in the fluid at the first boiling are killed, it is maintained at a temperature varying between 89° and 90° Fahr., which will induce any that may have escaped to advance to that stage of germination at which destruction would be certain at the second boiling. After this it is kept for some weeks again at the medium temperature, still secluded by the cotton-wool; if it remains clear and limpid for that time, it is fit for sowing, but if it becomes turbid, it shows that it has been imperfectly sterilised, and is unfit for use. If every precaution has been taken, the fluid will remain perfectly limpid for months, though the cultivation-tubes which contain it are only closed by some loose cotton-wool. Instead of cotton-wool, the

tube is sometimes closed by softening its neck by the blow-pipe, and drawing it out to a very small extremity, at the same time bending it to and fro in a number of sinuous twists. At each of these bends a little liquor condenses, which detains any impurities that might be carried in by the air. When the cultivation fluid has been inoculated with the virus, the same care is exercised in excluding these impurities, and it is kept at the temperature most favourable for the growth of the germs, which soon multiply so rapidly as to make it quite opaque in a short time.

With regard to the inoculation of animals, the prepared fluid is contained in closely corked tubes, those of Pasteur holding

Fig. 9.—*Tube containing the so-called "Vaccine."*



sufficient to inoculate 50, 100, 200, or 300 sheep; and they are labelled as "first vaccine," or "second vaccine," according to the degree of cultivation. Of course, it is most important that the liquid should be perfectly pure when it is used, as if not, inflammation of the skin, blood-poisoning, &c., might occur, and the object of the inoculation be frustrated. The tubes should be kept in a cool place, as in a cellar, and a tube which is opened one day should not be kept for use afterwards, but if any of the contents is left, it should be destroyed. Inoculation is made with the ordinary hypodermic or Pravaz syringe, such as is used by medical men and veterinary surgeons. This instrument should

Fig. 10.—*Hypodermic or Pravaz Syringe for Inoculation.*

be perfectly clean and free from germs every time it is used, and in filling it, care must be taken that air is not sucked in with the fluid to be injected. The piston should work so well as to completely fill the syringe. If sheep are to be inoculated, an assistant seizes the animal, seating it on the ground and holding it by the fore legs, while the operator inserts the needle of the syringe through the thin skin inside the thigh. When completely through the skin, the piston is pushed until sufficient fluid has been introduced—the body or piston of the syringe being graduated by marks, the quantity can be measured. The needle is

then withdrawn and the skin lightly rubbed, in order to diffuse the injected material beneath it. The syringe will hold fluid sufficient to inoculate eight sheep or goats, or four horses or cows; and with a little practice a man can operate on 150 sheep per hour. In twelve or fourteen days after the first inoculation, the operation is repeated with the second or stronger fluid, the opposite thigh being selected if sheep are the subjects.

Fig. 11.—*Inoculating a Sheep with the Virus.*

With the larger animals, the inoculation is made behind the shoulder in cows and oxen, and in the neck of horses. The skin being thick, care should be taken not to bruise it in pushing the needle through. This is best done by taking up a fold of the skin and passing the needle into it, the needle being stronger than that for sheep. If the first inoculation is not properly done, there is danger that the second may kill. The appearance of a tumour at the point of inoculation in cows has been rare, but in horses, and more especially young horses, this has been noticed; but it has generally subsided without any treatment.

The experiment of inoculating young horses three times—

twice with the first vaccine, and once with the second—has resulted favourably, no tumour appearing. This may prove to be the best method of inoculation for horses.

It may be interesting here to remark that, whether moved thereto by Pasteur's success in protecting poultry from fowl-cholera, or inspired by some other influence, there were other investigators of this disease in the field, who came near the goal which was subsequently reached by the indefatigable master. In 1880, Dr. Toussaint, of the Toulouse Veterinary School, published the results of his inquiries and experiments, and we learn that for nearly four years he had been almost exclusively occupied with the study of anthrax, and especially practising experimental inoculation of its virus. These experiments led him to the discovery that sheep and rabbits were protected by injecting beneath the skin anthrax blood deprived, he believed, of the bacilli, either by filtration, heat, or even antiseptics. He preferred heating fresh blood from a diseased animal to a temperature of 131° Fahr. for ten minutes, adding to it one-half per cent. of carbolic acid. A small quantity of this injected beneath the skin, gave rise to slight fever in three or four days, but which subsided on the fifth day. Inoculation with very virulent anthrax blood on the fifteenth day afterwards, produced no effect; so that the animal was proof against it. When injected at the same time as the heated material, however, death ensued.

Toussaint had, therefore, the priority of two years in discovering a means of securing immunity from anthrax, in modifying its virus by heat; but, unfortunately, his simple method has not been found to possess the advantages or safety of Pasteur's cultivations, though Chauveau has endeavoured to improve it.

Very early in 1878, Dr. Burdon Sanderson, the Superintendent of the Brown Institution, having received a money grant from the Royal Agricultural Society, undertook some experiments in anthrax ('Journal' of the R. A. Society, vol. xvi., p. 257), in which he succeeded in rendering cattle proof against the lethal inoculation of virus, by first passing the latter through the body of a guinea-pig, which it had killed. "The rodents die, but the bovine animals inoculated with their blood or with the pulp of their diseased spleens, recover." And he adds: "The question whether this fact, like the analogous one of the mitigation of human small-pox by transmission, can be directly applied to a practical purpose, I leave to be determined by future inquiry. Its present interest lies in its bearing on the nature of the process of infection in anthrax." In 1879 and 1880, the experiments were continued by Sanderson's successor, Dr. Greenfield, and

those who read his reports in the Society's 'Journal' (vol. xvi., p. 273, vol. xvii., p. 30), will find that Pasteur's discovery of anthrax protective inoculation by means of cultivated virus, was nearly, if not quite, anticipated by the liberal action of the Society and the scientific ability of its agent. Dr. Greenfield, in his second report, published at the commencement of 1881, concisely gives the object and the result of his investigations, from which it will be seen that, so far as immunity from the disease is concerned, they were the same as those of Pasteur.

"In the course of these experiments, it has been my endeavour to ascertain whether any modification of the poison of the disease could be artificially produced without the employment of animals for the production of the modified virus. With this object, I have made a series of experiments, cultivating the virus artificially in organic fluids, and I have been able to prove that it is capable of gradual attenuation, even to the degree of complete destruction of its virulence, and it may thus be greatly modified, so that a certain degree of poisonous activity may be attained at will." The details of the experiments given, quite corroborate his statement. Unfortunately, we have no Academy of Sciences in this country; and though our agriculture has been most terribly damaged by the ravages of pestilential diseases among animals, yet little attention has been paid to them. Consequently, the Society's efforts and Dr. Greenfield's anticipatory discovery, have, it must be confessed with regret, been allowed to drop out of sight. If we compare England with France, or some other countries, in this matter, we have small cause to congratulate ourselves.

The brilliant success attending Pasteur's public demonstrations in his own land, naturally attracted great attention in other continental countries which suffer quite as much loss as France from the destructiveness of anthrax, and some of these were eager to satisfy themselves as to the efficacy of inoculation, by a trial on their own territory. Hungary made a request, through Baron Kemeny, Minister of Agriculture, for M. Pasteur to make an experiment at Buda-Pesth; this was complied with, M. Thuillier, one of Pasteur's assistants, and who has since died from cholera while investigating that disease in Egypt, having been sent to the capital of Hungary to conduct the official trial in September 1881. A Commission was appointed to watch the experiment, Baron Kemeny being President, with the chief professors of the Veterinary Institute and of the Faculty of Medicine as members. Sixty sheep and ten bovines were provided, but some of the animals were weakly, and others were suffering from various diseases not apparent before inoculation. One-half of the sheep and one-half of the cattle were set aside

as test animals, to be inoculated with the crude virus only. Two of the sheep inoculated with the cultivated virus died from non-specific disease before the final test. When all were treated at last with the deadly virus, the efficacy of "vaccination" was fully verified by the resulting deaths in those not protected, and the immunity of those vaccinated. The Buda-Pesth experiment was repeated at Kapuvar on one hundred sheep and twenty oxen; fifty of the former and fourteen of the latter being "vaccinated," and fifty sheep and six oxen being reserved for tests. Fifty-nine sheep and one test cow died after virulent inoculation, and the surviving sheep and three of the cows were very seriously ill. One vaccinated sheep died, the other sheep and oxen not being affected. Twenty-six sheep of a flock ravaged by anthrax were also protectively inoculated. After the second inoculation ten of these died, and nine again subsequently.

This experiment was considered unsatisfactory, and the reason for its being so was explained by Pasteur as being due to the first inoculation material, which Thuillier took with him from France, being too weak to resist the effect of the second—it had been, unknown to Pasteur, "over-cultivated."

Prussia made application for "vaccine virus," but as the value of Pasteur's discovery was formally contested in Prussia, by Koch and others, Pasteur decided that it would be better to have a demonstrative experiment like that which had been made at Pouilly-le-Fort, Melun. Dr. Roloff, director of the Berlin Veterinary School, took the initiative in this step, by applying to the German Minister of Agriculture to have the experiment made, and to nominate a Commission to report on it. This was agreed to, and the Commission—with H. Beyer, Member of the Superior Council of Government, as President, and Dr. Virchow, Count Zieten-Schwerin, Councillors Zimmerman and Rimpau, several Veterinary School directors—Dammaun, Leisering, and Seidamgrotzky, and several of the leading veterinary surgeons from different parts of Germany, members. Twelve cattle and fifty sheep were collected at a place called Packish, where anthrax had prevailed to a large extent for some time, attacking horses, cattle, and sheep. In one year alone, seven horses, sixty-two cattle, and twenty-two sheep died; and it is to be noted that the outbreaks were more frequent when the animals grazed on certain parts of the field, or when these parts were mown for stall-feeding. The farmer had used this field as the burial-place for animals which had died from anthrax, while other carcasses he put in the manure-heap, which was in due course distributed over the land. The cattle and sheep for experiment were brought from an infected locality, and were divided in two equal portions; and twenty-five sheep and six

cattle were inoculated with the primary vaccine without any unfavourable symptoms following. After the second inoculation, however, several of the cattle and sheep became very ill, and three sheep died. The cattle and remaining sheep were soon quite well, and were inoculated with very virulent blood, at the same time as the other half of the animals kept for test purposes. All of the former continued perfectly well, but all the sheep and three cattle of the latter perished from anthrax. The experiment demonstrated that Pasteur's protective inoculation gives the inoculated animals complete immunity; but its practical utility was doubtful, as 12 per cent. of those inoculated succumbed to the second inoculation. Pasteur was of opinion that the breed of sheep had something to do with this result, so different to that observed in France; and the Minister of Agriculture, accepting this view, put the entire flock of sheep on the farm at his disposal. These were subjected to experiment; and fifty-two head of cattle on another farm were inoculated protectively. None of the cattle died; and the result with the sheep was more favourable than that in the first experiment.

The conclusions or inferences arrived at from these two sets of carefully conducted and closely supervised experiments, were to the following effect:—

"1. Cattle which withstand inoculation with the cultivated material, do not contract the natural form of the disease.

"2. The weakest cultivated fluid (*premier vaccin*) did not produce any sign of disease, either in cattle or sheep.

"3. The second inoculation, with the more virulent cultivated material (*deuxième vaccin*), produced a decided rise of the internal temperature and illness, although this was not, as a rule, fatal. At the seat of inoculation, various-sized nodules or swellings formed, which were either painful or painless. None of the cattle died after the second inoculation, but three sheep of the twenty-five used for the first experiment, and one of the 251 of the second, perished.

"4. The controlling experiments with the blood of an animal that had died of anthrax, and with the virulent fluid from Pasteur's laboratory, produced only a slight disturbance in the cattle and sheep that had been previously protected by the cultivated fluids; those animals which had not been so protected, however, became very ill indeed. Of the twenty-five sheep in the first experiment, and which had been inoculated with the protective fluid, none died; of the twenty-four in the second experiment, two lambs succumbed—one fourteen days after inoculation; but both were weakly animals, which may account for this unfavourable result."

From this it will be seen that the report of the Commission, to which Germany seems to have attached so much importance—judging from its composition—was entirely in favour of Pasteur's discovery; and it was concluded that on large farms where anthrax is more or less prevalent, it would be advantageous and economical to protect the animals (*herbivora*) by inoculation, especially the cattle, as it is not so dangerous for them as sheep, while they are more valuable.

It should be stated that all the sheep and cattle remaining from the experiments, and which had been protected, had been turned out to pasture upon the infected farm, and seven months afterwards they were still in good health; though some of the lambs born from the ewes subsequent to the experiments died from anthrax.

There can be no doubt that in countries or localities where the malady prevails, protective inoculation, when properly practised, must prove an immense boon. In India, for instance, which is in the unenviable position of having nearly, if not altogether, every contagious disorder to which animals are liable—cattle-plague, foot-and-mouth disease, sheep-pox, glanders, rabies, anthrax, &c.—causing a loss, at the lowest estimate, of six million pounds sterling a-year, and with scarcely any attempt at a remedy—protective inoculation for the prevention of anthrax among horses alone would prove of great benefit to our mounted corps there. And what would it not save if it could be applied to all these devastating plagues of animals which revel almost unchecked all over India? Already the possibility of obtaining such benefits have been shown by Mr. Mills, of the Army Veterinary Department, who at Madras, in 1884, conducted a series of experiments on ponies, donkeys, cows, bullocks, buffaloes, sheep, and guinea-pigs—in all 88 animals, with marked success. And Mr. Frost, of the same Department, appears to have been equally successful in Burmah; so much so, that the agents of the Bombay-Burmah Trading Corporation have expressed their intention of having their elephants in the Mingyan and Chindwin Valley forests inoculated against attacks of anthrax, which is very destructive to these creatures in that country.

Some parts, and often the best, the fertile *vleys* or valleys of South Africa, are deadly localities for horses at certain seasons of the year, because of the dread *Paard Ziekte* (horse-sickness). That this is a germ disease there can be no doubt whatever; because a horse that has recovered from it, which is rare, is believed to be safe from another attack—is what is called "salted," and is therefore very much more valuable than one which has not had the disorder. Besides, the germ which causes it has been identified, and successfully inoculated in guinea-pigs.

The disease is of the nature of, if not identical with, anthrax; and the Pasteurian inoculation would, it may safely be predicted, protect all the South African horses in the places where they would perish wholesale, if exposed.

In an immense empire like our own, the greater portion of which is agricultural, and nearly every part of which is haunted and harassed by contagious diseases among the animals useful to man, and so essential to his welfare, this method of guarding against disorders which are generally fatal, and rarely amenable to medical treatment, should have wide and most beneficial application, and especially when it can be resorted to for all of these maladies.

But to return to Pasteur's researches in anthrax.

Still pursuing his investigations with regard to that disease, and fowl-cholera, and having subdued the virulence or microbe of these to his will, bringing them down to a point where they could not multiply in the bodies of animals inoculated with them, and still keeping these organisms alive in suitable media, he demonstrated that these now non-virulent microbes could be restored to their pristine activity, and rendered capable of living and multiplying in the blood of animals. An attenuated anthrax virus which would not affect a guinea-pig even a week old, will yet kill one just born, or one or two days' old; and if some of the blood of this is inoculated into an older one, this also will perish; while should the blood of this, again, be injected into a still older one, this will succumb; and so on through a series, until at length the blood has acquired such power that the old guinea-pigs, sheep, and finally oxen, are unable to resist the smallest drop of it. It is the same with fowl-cholera. The most attenuated and harmless cultivation of the microbe can be made as deadly as ever, by passing it successively through small birds and chicken, until it is capable of destroying full-grown poultry.

The attenuation of the poison of contagious diseases by the air, suggested to Pasteur the possibility of the great epidemics of mankind (and for that matter also, the epizoötic diseases of animals), being modified or extinguished, and reappear or become more intense, through the same agency. "The accounts which I have read of the spontaneous appearance of the plague in Benghazi in 1856 and 1858," he says, "tend to prove that this outbreak could not be traced to any original contagion. Let us suppose, guided by the facts now known to us, that the plague, a malignant disease peculiar to certain countries, has germs of long duration. In all these countries its attenuated virus must exist, ready to resume its active form whenever the conditions of climate, of famine, or of misery present themselves afresh. The

condition of long duration in the vitality of the germs is not even indispensable; for, if I may believe the doctors who have visited these countries, in all places liable to the plague, and in the intervals of the great outbreaks of the epidemic, cases may be met with of people attacked with boils, not fatal, but resembling those of the deadly plague. Is it not probable that these boils contain an attenuated virus of this disease, and that the passage of this virus into exhausted bodies, which abound only too freely in periods of famine, may restore to it a greater virulence? The same may be the case with other maladies which appear suddenly, like typhus in armies and in camps. Without doubt, the germs which are the cause of these diseases are everywhere scattered around, but attenuated; and in this state a man may carry them about with him, or in his intestines, without suffering much, if any inconvenience. They only become dangerous when, through overcrowding, and perhaps successive developments of them on the surfaces of wounds, in bodies enfeebled by disease, their virulence becomes exaggerated."

The manner in which animals became infected with anthrax also engaged Pasteur's attention, now that it had been definitely established that it was due solely to a microbe or its spores. This organism is now well known to all pathologists as a rod-shaped body, multiplying, when allowed oxygen or air, by dividing across, and measuring from $\frac{1}{1250}$ to the $\frac{1}{2500}$ of an inch in length, and about $\frac{1}{1800}$ of an inch in breadth. It has an outer case or sheath, and inside this is fine granular protoplasm, the vital part of the organism. In this the spores appear as glistening points, which gradually become ovoid, and increase in size by the protoplasm collecting closely around them, until they fill the rod or sheath, like peas in their pod; the sheath now either breaks off in segments, which burst, and the spores are set free; or it becomes softened and dissolves, leaving them at liberty in the middle of the pulp. These, as has been already mentioned, are extremely tenacious of vitality, and develop into the rod-like bodies or bacilli, when cultivated artificially, or introduced into the blood of animals.

When sheep, cattle, or horses at large become affected with anthrax, this must be due to their receiving the spores from some external source; and that this may be the pasturage, was proved by an experiment, in which several groups of sheep were fed on grass which had been sprinkled with artificially-grown bacilli or their spores. Some died from anthrax, and many survived after having been visibly affected—the period of latency being in some cases eight or ten days, in others much less. If prickly or stubbly plants, however, were added to the infected herbage, the

mortality was very much increased. Experiment also confirmed the view to which this artificially infected herbage gave rise, that pastures become tainted by the excreta, which are often bloody, of living diseased animals, and by the blood and *débris* of those which have died or been killed, and buried on the spot. Adding some anthrax-blood to earth which had been sprinkled with yeast-water or urine, and keeping this at summer temperature, or at that which the fermentation of a dead body sets up around it—as in a dung-heap—the bacilli of the blood had, in less than twenty-four hours, multiplied and developed spores. Afterwards these spores were observed in their latent or resting condition, ready to germinate and produce the disease in animals, perhaps years afterwards.

To ascertain what really happens in ordinary circumstances, the earth from a grave in which a sheep that had died of anthrax from natural infection, and the carcass of which had been inspected by Pasteur and his assistants, was examined ten and fourteen months after that event, and was found to contain spores of the bacillus. Guinea-pigs inoculated with these spores died from anthrax. The same experiment was successfully made with soil from the surface of the grave which had not been disturbed since the burial of the sheep.

Experiments were carried out with earth obtained from trenches, in which the carcasses of cows dead of the disease had been interred at a depth of more than six feet two years before. Material was procured from this soil, which at once produced the disorder. On three occasions this happened, though nothing of the kind was exhibited in the earth from around these trenches. Notwithstanding the operations of ploughing, sowing, and reaping, the surface-soil of the graves still contained anthrax-germs. From the carcasses of the dead animals the earthworms bring these germs, as well as those of putrefaction and others, to the surface; and these otherwise useful creatures, themselves unaffected, become the indirect cause of the disease in animals which graze on such ground.

M. Tisserand, Director of Agriculture in France, has remarked that the malady is unknown in the Savarts of Champagne, although this locality is surrounded by districts which are notoriously infected; if it chances to appear, it is owing to its introduction from without. Here the arable soil is not more than four or five inches thick, and lies on chalk; so that it is unfavourable for worms, and therefore the germs in the bodies of dead animals will remain in them, and the soil will not be tainted. It is on soils of the opposite character that anthrax is most likely to prevail, if anthrax-infected bodies are buried in them. Impressed with this knowledge, Pasteur recommends that

such bodies should never be buried in fields intended for cultivation, forage, or for pasture. If possible, a sandy or poor calcareous soil, dry and readily desiccated, should be selected, as it is not suited for earthworms.

But with all due deference to M. Pasteur, I would urge that, whenever and wherever animals die from germ-diseases, they be not buried, but burned. This is the only safe procedure with contagious maladies, and particularly in countries with rich and heavy soils ; it is that which is resorted to by military veterinary surgeons in India, in disposing of the carcasses of horses which die from that form of anthrax known as "Loodiana Disease" so named from the country in Hindostan in which it was first noted as most prevalent.

While we are on this subject of anthrax, it should be mentioned that a disease which was, until three or four years ago, considered as one of its manifestations, is now distinguished as a different malady, but, like it, is due to a microbe capable of artificial cultivation, and is then protective from the disease when inoculated. I allude to a most fatal disorder of young stock with which cattle-breeders in this country and in our colonies are only too familiar, and which, among a variety of local names more or less peculiar or significant, is perhaps best known as "Black Quarter," but which is now technically designated "Carbuncular Emphysema, Erysipelas," or "Symptomatic Anthrax." It usually attacks the most rapidly thriving of young bovines, its onset being remarkably sudden, and its course strikingly rapid, the termination being almost invariably fatal. The chief symptom is sudden lameness—generally in a hind limb, swelling of the quarter from which, if the skin covering it is pressed upon, a crackling sound is emitted, due to the disengagement of gas from the large quantity of dark blood thrown out beneath, and which forms such a characteristic feature on dissection after death, as to give the disease its familiar name of "Black Quarter."

Incited thereto, probably by Pasteur's example, Arloing and Cornevin, Professors at the Lyons Veterinary School, and Thomas, a veterinary surgeon at Bassigny, France, have studied this malady, known as "Chabert's disease" in that country, and have demonstrated that it is very different from anthrax, in that it is not communicable by inoculation of the blood, which that disorder most certainly is ; that its microbe is like a large bacillus, but moves, and is only found in the affected limb among the serum, or deep among the muscles, to the fibres of which it adheres so closely that it can only be removed by scraping with a sharp knife ; and that this organism, when injected into a vein, causes only a slight and brief disturbance of health, whereas the

anthrax microbe would assuredly kill. Death is more rapid in this disease than in anthrax, and the spleen is not involved, as in it. But the most valuable part of their discovery lies in the fact they have established—that this direct (intravenous) injection safeguards the animals so operated upon from the mortal effects of the disease, either naturally or artificially, which occurs when the virus is introduced among the muscles. The protection has been found to last for more than two years; as cattle which had received the virus at that time were inoculated with a quantity of virulent muscle-pulp, but remained unaffected, while another bovine, which was unprotected, died on receiving one-tenth part of the dose. The virus does not seem to be capable of attenuation by cultivation, and its introduction into the blood through a vein is the most effective, and hitherto the most simple, means arrived at to save cattle exposed to infection from death.

This method, however, demands careful manipulation to prevent the inoculating material from escaping into the space between the skin and the vein, where it would set up the disease itself, and speedily destroy the animal.

It may be remembered that, a few years ago, Dr. Burdon Sanderson undertook some experiments for the Royal Agricultural Society, in contagious pleuro-pneumonia of cattle, in which he sought to ensure protection by introducing the virus through a vein into the blood, instead of beneath the skin of the tail, as in the ordinary procedure. And as with "Black Quarter," so with this disease. The intravenous injection of the virus appeared to ensure perfect immunity against the lung disease; but there was great danger lest any of the virus should get beneath the skin during the operation, as it would give rise to so much local and general disturbance as in all likelihood to cause death. The microbes of the two diseases, while producing very little derangement to the health when imported directly into the blood stream, though bringing about such changes as will prevent attacks of their special maladies, yet find such a congenial *habitat* in the connective tissue immediately beneath the skin, as to multiply in appalling numbers within almost a few hours, and thus to destroy life. The safety of tail-inoculation for Lung Plague is probably to be found in the very small amount of connective tissue in that part, though the operation is no less certain in its results, so far as protection is concerned.

Pasteur has given his attention to another disease with which agriculturists, and pig-breeders and feeders, are only too well acquainted. I allude to Swine-plague, known in France as "Rouget," and in Germany as "Rothlauf." This malady has for some years assumed such a virulent and deadly character,

and has attained such widespread dimensions in Europe and the United States of America, as to threaten the extinction of the porcine tribe within a brief space, if prompt sanitary or protective measures are not adopted. From its superficial resemblance to anthrax, it was long looked upon as a form of that disease; but by Dr. Budd and a few veterinary surgeons, it was considered to be analogous to typhoid fever in man, and was consequently designated "Pig Typhoid." In August, 1875, having had the opportunity, while with the Royal Engineers at Chatham, for the first time of examining some pigs that had died of this disease at Higham, near Gravesend, I came to the conclusion that it was neither anthrax nor typhoid fever, but a distinct disease of the swine tribe.* Some time after this (1877), Dr. Klein came to the same conclusion, and others were subsequently of this opinion; so that it is now decided that it is a disease peculiar to the pig. So far as experiments have shown,

Fig. 12. *The Organisms or Micrococci of Swine-plague.*

it can only be transmitted by inoculation to the rabbit, mice, and white rats, among mammalia; and among birds, according to Pasteur and Thuillier, Cornevin, and some other French investigators, the pigeon can be infected—very readily, say the first-named authorities,—but Klein denies this receptivity in pigeons. It is most infectious and contagious among pigs, in which it appears in two forms:—acute and chronic; and its maintenance is due to an ærobic germ, different from that of anthrax in being in the form of minute round granules or "cocci," single or double, as in a figure of 8, slightly motile, isolated, or collected in groups of two, three, or four. These are easily cultivated in the same

* "A Fatal Eruptive Fever in Pigs."—'Veterinary Journal,' October, 1875, p. 269.

manner as the anthrax rods or spores. The discovery of the microbe was made by Detmers in America (1882), and about the same time by Pasteur and Thuillier in France. Klein had previously found a bacillus, but this is not now acknowledged as the active agent. The germ exists throughout the body of the diseased pig, and especially in its *excreta*, which are very virulent. Infection is easy by the digestive organs; and, besides other modes of contamination, the food may receive the microbes by diseased mice, or through the medium of rats, flies, birds, &c.

In 1883, Pasteur made known a method of attenuating the virus, which, simple and effective, offers as certain protection against swine-plague, when inoculation is properly performed, as anthrax inoculation does against that malady. He found that when a pigeon was inoculated in the chest with the microbes of the disease, it died in six or eight days. When a second pigeon was inoculated from this, a third from it, and so on in succession, the microbe acclimatised itself in pigeons; so that death ensued more rapidly, and the blood of the last pigeon assumed much more virulence when introduced into pigs, than the most infectious products of a pig that had died of the natural disease. But the passage of the microbe through rabbits had quite a different result. When inoculated from a diseased pig, these creatures all speedily perished, but cultures of their blood in sterilised media became progressively easy and more abundant, the microbe somewhat changing in appearance. When pigs were inoculated with the blood of the last rabbits, and the results were compared with those obtained from the first, it was discovered that the virus had gradually lost its power; and though the blood of these rabbits made pigs very ill, it ceased to kill them. On recovery, they were proof against the fatal disease.

In various parts of France, this swine vaccination has been carried out; but perhaps the most carefully conducted and extensive experiments were carried out in the Grand Duchy of Baden, in 1884, to test the value of the measure. In that country swine-plague had caused heavy losses for many years, and in 1865 and 1871 commissions were appointed to report upon it. In 1884 it was arranged that a trial of vaccination should be made, under Pasteur's direction, the experiments to be carried out by one of his assistants, and the inoculating material and instruments to be supplied from his laboratory. The experiments were carried out at fifteen places on 237 pigs of different breeds and ages, and were under the observation of selected veterinary surgeons from Berlin, Munich, Stuttgart, Hesse, Alsace-Lorraine, Luxemburg, and Berne, as well as others. All the pigs were in perfect health, and the inoculations were performed in a similar manner to those on the sheep

at Berlin. One-half the number were kept as test animals, and were not protectively inoculated. The second vaccination was made twelve days after the first, no ill effects following, except to six animals ; and the final test was applied to all the pigs, in another twelve days, either by inoculating them with the deadly virus, or by feeding them on food mixed with it. The experiment was as conclusive as that at Pouilly-le-Fort, with regard to anthrax. The pigs which had been protectively inoculated were unaffected by the crucial test, while those not so indemnified nearly all succumbed to the natural or imposed infection.

Rabies in animals is a disease which concerns people in general, from its terrible character when communicated to mankind as Hydrophobia ; but to agriculturists it possesses additional interest, because, in addition to the danger they personally incur, the losses they are liable to sustain among their stock from the bites of rabid dogs are by no means insignificant at times, in localities where the scourge prevails.

Pasteur, busy in his laboratory investigating different diseases of the human species—typhoid fever, a serious bone-disorder, boils, puerperal fever in woman—in all of which he found special organisms, whose action he endeavoured to annul, applied himself more especially to the study of hydrophobia, which, for a long time, had occupied his mind. He had been particularly struck by the peculiar view entertained by many physicians, and others, that this fatal disease was largely, if not altogether, due to mental excitement, and that contagion had little if anything to do with it. Of course, these physicians were not acquainted with veterinary medicine, and had no experience of rabies in animals, else their imagination could not have so misled them. An opportunity occurred for him to collect some of the mucus from the throat of a child a few hours dead from the malady. With this he inoculated some rabbits beneath the skin, and they died in thirty-six hours ; their saliva conveyed the disorder to other rabbits. There was nothing at all novel in this, as rabbits had been infected through experimental inoculation years before. But death had been so rapid in Pasteur's rabbits, that there was no time for them to have had the disease. More rabbits were inoculated, and in the blood and tissues of those dying and dead he found a special microbe, which was easily cultivated in sterile infusions, and produced death in other rabbits, reappearing in them. This organism was also discovered in the saliva of children suffering from other maladies, and was not therefore special to hydrophobia. Grown in successive cultivations at twelve hours' interval, its virulence remained undiminished ; but if allowed to

remain longer in contact with the air it became enfeebled, and attenuated cultures could thus be obtained; these inoculated in other rabbits were found to prevent the action of the strong virus. Though both ærobic and anærobic, the air destroyed its potency very quickly; but kept from the air, it retained its deadly character for months. It is much to be regretted that Pasteur has not stated whether he tried dogs with this microbe; as its discovery, it must be acknowledged, did not advance the question as to the exact cause of rabies.

The incubation of rabies is so uncertain, and sometimes so protracted, when produced in the ordinary way, that Pasteur became tired of waiting for months for the results of his inoculations, and therefore sought a more certain and rapid means of developing it. At length he could make it appear by introducing portions of the brain and spinal cord of dead rabid animals beneath the skin of dogs and rabbits, and this nervous matter could be kept in a state of purity for a long time; whereas rabific saliva was always impure, and lost its potency in twenty-four hours.

A happy thought induced Pasteur to place this pure nerve-virus on the surface of the brain of a dog, by trephining its skull while it was under the influence of chloroform, and it became rabid in a few days. Other dogs were treated in the same way, and nearly all yielded to the malady in less than twenty days. This showed that in the nerve substance the poison of rabies exists in its most concentrated form, and especially in the brain and upper part of the spinal cord (medulla oblongata)—the microbe of the disease evidently flourishing there most luxuriantly; though the virus is also localised in certain salivary glands and mucous surfaces; all of which peculiarities may account for differences in symptoms in cases of hydrophobia. It was ascertained that the disease could be developed in an animal by passing infected brain-matter into its blood, through a vein, almost as quickly as by trephining.

For a number of years, in this country and on the Continent, by means of the microscope, extremely minute round bodies have been noticed in and around the upper part of the spinal cord and brain of people and animals dead from the disease; these were also observed in Pasteur's laboratory, and it is not at all improbable that they are the germs of rabies.

When the virus was passed through a certain series of monkeys, it gradually lost its power, until it was so enfeebled that dogs inoculated with it did not suffer from rabies, but were, on the contrary, rendered insusceptible to rabific inoculation. By such means Pasteur succeeded in rendering some sixteen out of every twenty dogs he inoculated refractory to rabies.

But remarkable and important though this result might be considered, he became impatient at having to wait for months before he could assure himself that such animals were really rendered proof. He therefore set about to find a method by which he could produce viruses of different degrees of strength, so that he might obtain more certain, more prompt, and more practical results.

This he at last effected by inoculating a rabbit on the brain with a portion of the spinal marrow of a rabid dog—this caused the disease to appear in about fifteen days. The virus was passed on to a second rabbit in the same way, and from this through a number—twenty to twenty-five—when the interval of latency was reduced to seven days. This happened with the greatest regularity in the experiments; and though the inoculations from rabbit to rabbit were carried up to ninety, the incubation period could not be reduced much below seven days. It may be mentioned that the experiments were carried on for more than three years without interruption, and without having recourse to any other virus than that of the successively infected rabbits.

Consequently, nothing was easier than to have constantly on hand a supply of rabific virus, perfectly pure, and always of the same quality or potency; and this was the practical feature of this long and laborious inquiry into the method of inoculation. The spinal cord of these rabbits was equally virulent throughout its whole extent, and if slices from it were removed, with all precautions to maintain their purity and freedom from putrefactive germs, and suspended in dry air in flasks (air dried by placing fragments of caustic potass at the bottom of the vessels), their virulence slowly disappeared until it vanished altogether—the rate of disappearance depending upon the thickness of the slice, each spinal cord being cut up into fourteen or fifteen pieces. Temperature also influenced its disappearance; as the lower this was, so the longer was the virulence maintained. If kept in carbonic acid gas, in a humid condition, air being excluded and no foreign microbes admitted, the pieces of nerve-tissue would remain potent for at least several months. These results constitute the scientific feature of the method.

When it was desired to render a dog refractory to rabies in a relatively brief period, a fragment of fresh rabific spinal cord from a rabbit dead seven days after inoculation, was suspended every day in a series of flasks, the air in which was dried in the manner above mentioned. Every day also the dog was inoculated beneath the skin with a morsel of this dried spinal cord mixed up in a little sterilised broth—commencing with

the oldest fragment, to make sure that it was not too virulent; each day a more recent fragment (an interval of two days' drying between each piece), until the last was reached, and which was very virulent, being only one or two days in the flask. After this the animal could be inoculated beneath the skin or on the surface of the brain with the most active virus, and rabies would not follow.

Pasteur had inoculated fifty dogs of all ages and breeds, and rendered every one of them proof against the disease, when, unexpectedly, on July 6th, 1885, three individuals from Alsace arrived at his laboratory; two of them brought a boy nine years old, said to have been worried by a rabid dog on the 4th of that month, and the wounds, fourteen in number, gave evidence of this. Two professors of medicine saw the boy on the same day as Pasteur, and from the history of the case and the nature of the injuries, it was considered a desperate one—death from hydrophobia appearing inevitable.

Pasteur therefore decided, though not, it may be imagined, without grave misgivings, to practise on this child the method of protection which had invariably succeeded with dogs, even *after* they had been bitten by rabid ones. The step was a terribly venturesome one, and nothing but the apparently hopeless position of the patient, and the confidence in his method begotten by uniform success, could have endowed him with courage, or inspired him with hope, in undertaking by far the most formidable and most anxious task of his lifetime.

The disease is horribly agonising and dreadful—more so, perhaps, than any other which afflicts humanity; and while Pasteur was naturally anxious to save the boy from it, the risk was fearfully great that the very means employed to do this might produce it.

On the evening of July 6th—sixty hours after he was wounded—the boy received the first subcutaneous injection of spinal marrow, obtained from a rabid rabbit fifteen days before, and kept since then in a flask of dry air. The following days' inoculations were made in the same region—beneath the skin of the abdomen; so that thirteen inoculations were given in ten days, though Pasteur believed that a less number would have sufficed. The pieces of spinal cord had been dried from fourteen days to one day; and inoculations on trephined rabbits, made at the same time, showed that the five oldest specimens were not virulent, because they did not induce rabies; while the five subsequently dried ones were all active, their virulence being proportionally greater as they were more recent, until the last killed in seven days. During the latter days, therefore, the child

had received, successively, quantities of rabific matter, the last of which was so deadly as to destroy rabbits in seven days and dogs in ten—matter more virulent even than the poison of the rabid dog, because it had become intensified in its passage through the long series of rabbits.

This extreme virulence, though it might give rise to the gravest apprehensions, so far as the safety of the unfortunate child was concerned, yet had the advantage of limiting the duration of these apprehensions; as if it produced the disease at all, it must do so sooner than the ordinary dog virus would. But up to the time of writing this paper, the child remains unharmed—a living testimony to one of the most wonderful and valuable discoveries in medicine of this age, or perhaps of any time: a discovery vastly greater than Jenner's vaccination, great as that was.

Commencing with the discovery and the mastery over the microbes which destroy the stability of simple organic compounds and cause fermentation, by consistent reasoning, persistent experimentation, and profound — almost instinctive— powers of discernment, Pasteur advanced step by step along the same path, until he controlled, we may say vanquished, that form of fermentation, also due to micro-organisms, which destroys the life of man and beast. The most intense virus introduced into the body, either naturally or experimentally, had no power whatever for mischief after his protective inoculations; indeed, it seemed rather to consolidate and confirm the refractory condition with which these inoculations had endowed the individuals so protected.

It may well be imagined that Pasteur's boldness in venturing from animals to mankind, in his attacks upon disease, startled the medical world and the public more than anything he had yet attempted, and this all the more when the malady he dared to grapple with was hydrophobia—the mere mention of which seems to conjure up feelings of dread and panic. By many, the successful issue of the inoculations which the first patient had undergone, and that of the large number of people submitted to them subsequently, was received as a sure guarantee of immunity, even after several days had elapsed since injury was inflicted by rabid dogs. For Pasteur had proclaimed that, though a certain number of days (thirty) might have transpired after a person had presumably received infection from a mad dog, yet his attenuated virus, administered in the way described, anticipated the action of the slower-operating poison, and neutralised it. But in dealing with this, the most uncertain and hitherto intractable of all maladies, it was perfectly reasonable that many

who were not at all prejudiced against the wizard of the Rue d'Ulm, but were, on the contrary, his enthusiastic admirers, should hesitate before accepting this latest and grandest conquest as absolutely demonstrative of his power over such a disease. The oftentimes very protracted period of latency or incubation, and the uncertainty of inoculation from rabid dog-bites—only a comparatively small proportion of those wounded suffering subsequently—caused many who knew the disease to withhold their applause until they saw whether the results of the experiments on animals could be applied to man with the same certainty and safety. But so far as matters have progressed up to the present time, there is every reason to believe that the most sceptical will soon have reason to acknowledge the genuineness of this splendid triumph in prophylactic medicine.

On March 1st of this year, Pasteur announced to the Academy of Sciences that he had inoculated no fewer than 358 persons, many of whom had been bitten by really rabid dogs, and the others by dogs suspectedly rabid.* Of these people, 100 were bitten before December 15th, and the remainder subsequently; yet all, except one, had remained in good health, even the operation of inoculation being unaccompanied by any inconvenience. The case which died was that of a child severely bitten by a rabid sheep-dog on October 3rd, but who was not brought to Pasteur until thirty-seven days afterwards, when inoculation was practised, though against Pasteur's inclination, as he only too correctly surmised that the delay was too protracted. The child died of hydrophobia. The inoculated persons have come from all parts of Europe, and some from America.

It is now proposed to establish a Pasteur Institute in Paris, for the reception of people who have been bitten by rabid animals, where they will be protectively inoculated against the disease.

It is difficult to arrive at a satisfactory interpretation of the method by which Pasteur renders animals and mankind impregnable to the infection of rabies. He is himself of opinion that the facts are not in accord with the notion that contact with dry air gradually diminishes the intensity of the virus in the spinal marrow until it becomes null; so that the method is based on the employment of a virus at first almost inert, and then increasing in potency. The lengthened duration of the latent stage is not due to enfeeblement of the intensity of the virus, but to its diminution in quantity. So that the immunity

* It is perhaps needless to add that the list of patients sent to Mons. Pasteur increases day by day.—Edit.

might be given by inoculating very small, but daily increasing, doses of the same very active poison.

Another explanation he gives, which appears rather strange, but which agrees with certain results observed in studying micro-organisms, and especially the pathogenic, or disease-producing, microbes. Many of these appear to produce, in their cultivating media, matters which possess the property of hindering their own development. Since 1880, Pasteur had been making researches with the view to establish the fact that the fowl-cholera microbe produces its own poison. He has not yet been able to demonstrate the presence of this matter, but he thinks this can be done by using pure carbonic acid gas. The microbe of swine-plague is cultivable in very diverse kinds of infusions, but its increase in weight is so small, and it is so promptly arrested in its proportion, that it does seem that something is elaborated which stops the growth of the organism, whether cultivated in air or in a vacuum. Professor Raulin, in 1870, announced that the mould—*Aspergillus niger*, when vegetating, develops a substance which partially checks its growth, should the nutritive medium not contain iron salts. It may be, therefore, that the rabific virus contains two distinct substances—one being living and capable of multiplying in the nervous system ; and the other, non-vital, having the faculty, when in certain proportion, of interrupting the development of the first.

Thus far I have attempted to trace, though feebly, I fear, the progress made by Pasteur, in his noble endeavour to benefit mankind, by devoting his body and mind incessantly to the study of part of a subsection of the organic world, consisting of organisms which, if infinitely mean in their dimensions, are yet amazingly powerful in their action, and of which little, if anything, was known before his advent. The record of his labours, and the success attending them, should be sufficient to stamp him as one of the great benefactors of the human race ; his last achievement as far transcending his previous victories, as his whole work must raise him above those, whose one or two discoveries have conferred immortality upon them. Though quietly and unostentatiously toiling for so many years without seeking for fame or reward, overtaxing health and strength, and incurring grave risks of infection from the poisons which he sought to render tractable and useful, as well as encountering disparagement, incredulity, and opposition, he has nevertheless risen to the highest pinnacle of greatness, and France has generously acknowledged how much he has added to her honour and her well-being.

Professor Huxley, some time ago, in alluding to the benefits

which have resulted from his labours, and pointing out the manner in which he has opened out such sources of wealth to industry and agriculture in his own country alone, has said: "Pasteur's discoveries suffice, of themselves, to cover the indemnity of five milliards of francs paid by France to Germany." But his discoveries in contagious diseases and their prevention, cannot be estimated at a money value; nor can the effect of these discoveries on present and future investigators be overlooked. The possibilities which his researches have thrown open, to my mind, are amazing, and have surprised Pasteur himself.

His discoveries he looked upon as only a mere beginning, and he has often said, "You will see how all this will grow in time. Would that my life were longer!" And all friends of science and humanity must heartily echo the wish.

Some may be inclined to ask what practical value Pasteur's "Vaccination" is likely to have in dealing with contagious diseases: and others may deny that the method will ever possess advantages over the ordinary methods of combating them. This question of practical utility is worthy of consideration, and until his vaccination has been tried extensively for some time, it would be somewhat imprudent to hazard an opinion one way or the other. We have no evidence that, in the enfeebling or attenuating of the virus of these disorders, the germs are positively destroyed. On the contrary, we know they are sufficiently active to remove from the blood and tissues the pabulum which would serve for the multiplication of another generation of their kind, or produce in the body they invade the hypothetical inhibitory substance which will act as a poison to them. And we also know that weakened germs can regain their pristine virulency when placed in favourable conditions. Better by far, then, to utterly destroy the germs, even should this necessitate the destruction of the animals whose bodies they have taken possession of for their breeding ground, than keep them alive in a semi-latent condition, from which they may start with all their lethal vigour whenever circumstances allow them.

The apparently barbarous and unscientific "stamping-out" process has this to commend it, that, if thoroughly carried through, it exterminates the germs, as pulling up and burning noxious weeds before they seed clears the land of them. Therefore, in special cases, as where the disease is not very widely spread, and can be eradicated in this way without heavy loss, "stamping-out" by slaughter—repulsive though it always must be—must be preferable to protective inoculation. But it is evident that such a summary method can never be attempted with mankind; and even with animals, under certain conditions,

it would be futile. Take such a malarious malady as the South African horse-sickness, for example, which is indigenous to the *Vleys*, or low-lying valleys, and is due to germs in the water or on the moist herbage, which horses eat at a certain season. Such a disorder cannot be extinguished by destroying the sick horses, as the germs infest the soil, the grass, the water, and perhaps the very air is laden with them at certain hours during the sickly period of the year. And so with other animal disorders of a contagious character, which are widespread, and so treacherous in their infectiveness and insidious in their progress, that the skill and vigilance of man cannot grapple with them. For these the benefits of protective inoculation may be invoked.

Daily the disciples of Pasteur enlarge the territory in which he was the first to break ground, and reap such a marvellous harvest; and the special microbe of each specific disease of man and beast is being revealed, and in some instances cultivated so as to serve as a defence against its uncultivated congener.

One of the most treacherous and invasive, as well as destructive diseases of cattle, which is sapping the vitality of the best

Fig. 13.—*From the Tuberculous Deposit in the Lung of a Cow.*

Showing two large or "giant-cells," and two small cells, containing the germs or bacilli of Tuberculosis. Magnified 700 times.—*Klein.*

breeds, and which is communicable from the bovine to some other species—most probably also to mankind—through consuming infective milk and flesh, is Tuberculosis, or "Consumption." The microbe, or bacillus, of this infectious scourge was first clearly demonstrated by Koch, though previously Toussaint had found a micro-organism somewhat unlike this, but which may have been another form of the bacillus. He also succeeded in producing a culture fluid, which, when inoculated, caused the disease. Gerlach, of the Berlin Veterinary School,

had also before this proved the ready transmissibility of tuberculosis, being corroborated by Toussaint, Chauveau, and others. But, unfortunately, this bacillus has not yet been made subservient to man in affording protection from consumption.

The bacillus of Glanders has been discovered and cultivated by Schutz and Löffler, but it does not yet ensure protection from that malignant pest of horses.

Micro-organisms (micrococci) were observed in Cattle-plague by Klebs, in 1872, and by Semmer, of the Dorpat Veterinary School, two years later. In 1882, the latter cultivated this microbe, and found it grew abundantly. A calf inoculated with the culture died in seven days from rinderpest. When transferred, the cultures gradually become weaker, and sheep inoculated with them do not perish, but are rendered refractory to infection. Semmer has also discovered the microbe of dog Distemper.

Fig. 14.—*Pus of a Pulmonary Abscess in a Horse dead of Glanders.*

1. The nuclei of pus cells. 2. The glanders-bacilli.
Magnifying power, 700.—*Klein.*

Lustig, of the Hanover School, has discovered a bacillus in horse Influenza, which he has grown and inoculated. The same authority has also described, and experimented with, the micro-organism of Lung-plague, which had already been discovered by two Professors at Louvain University. Not long ago the Agricultural Society of Pavia, alarmed by the havoc wrought by Foot-and-mouth disease, appointed a Committee to study its progress and nature. The Secretary was Dr. Nosotti, who verified the observation of the veterinary professor, Rivolta, that the disorder was due to a microbe—the *Micrococcus aphtosus*—inoculation of which always gave rise to the disease, which could be rendered benignant by cultivation of the germ. And still more recently, Dr. Klein has published a short paper on this organism. On feeding sheep with the twentieth-generation cultivation, he produced the disease in them.

I might further allude to similar discoveries, but this

reference will testify to the immense impetus Pasteur has given to the study of these so-called "germs,"—a study which has become so important as to constitute a section of biology, under the designation of Bacteriology.

There can be no doubt that the more we are acquainted with the nature, mode of development, vitality, and special peculiarities of these microscopic entities, so the better we shall be able to destroy them, utilise them, resist their lethal influence, or avoid them. When we know, for instance, that septic germs are, in all probability, the active agents in the production of "navel ill" in lambs, "joint ill" in foals, and "heaves" (inflammation of the womb) in ewes, we can readily imagine how these serious conditions may be prevented by the exercise of a little care. One cause of abortion in cows is, with great likelihood, laid to the charge of germs; and the so-called blood-poisoning, which so often follows surgical operations, injuries, and parturition (puerperal or parturient fever), is due to the malevolent living atoms, which seem to be omnipresent, and are ever eager to commence their natural course of proliferation. To the surgeon, to the veterinary surgeon, and also to the agriculturist who breeds, rears, or employs animals, this knowledge is of great moment. To the latter, also, it is of additional importance, seeing that the germs play a lively *rôle* in the dairy, in ensilage, in nitrification of soils, in the improvement or deterioration of food for animals, and in many other ways, which, until Pasteur began to throw light on them, were never suspected.

Errors are almost inevitable in such a study as this of Bacteriology, which is so difficult and intricate, and demands an excess of patience and powers of observation that are given to few. For it must be remembered that each of these micro-organisms has a clearly-defined part to play in the programme of Nature; each has a chemical and distinct individuality, though in some instances it may be within a narrow compass, and though the differential anatomical or structural peculiarities of each may be most difficult, if not impossible in some cases, to find out. They each have their affinities or predilections for nutriment, locality—whether in or out of the body—and their special mode of finding access to a suitable *habitat* and development there. Only with the most laborious care, and special management and processes, can some of them be rendered visible.

We may therefore conceive something of the magnitude of the task which investigators in this wide and only partially-explored region have to encounter, especially when we learn that as many as 50,000 of some of these bodies would be required to make up the bulk of a small cheese-mite. To have

accomplished so much and so thoroughly in this direction—to have been the pioneer in investigations which have already revolutionised medical doctrine, and greatly added to our knowledge of natural phenomena — to have effected large economies in important industries and agricultural operations ;—but, above all, to have given us the means of averting or resisting the most baneful and pestilent diseases, is the honour to which Pasteur is entitled, and which will be gratefully accorded him now, and in still larger measure hereafter.

LONDON: PRINTED BY WILLIAM CLOWES AND SONS, LIMITED, STAMFORD STREET AND CHARING CROSS.